

INTRODUCTION:

Asthma is defined as a chronic inflammatory disease of airways that is characterized by increased responsiveness of the tracheo-bronchial tree to a multiplicity of stimuli¹.

The common risk factors for asthma symptoms include urbanization, airway irritants (pollen, animal, hair, industrial smoke, vehicle exhaust fumes, dust mites in pillows and bed covers etc)².

There has been a sharp increase in the global prevalence morbidity, mortality and economic burden associated with asthma over the last 40 years. Approximately 300 million people worldwide currently have asthma and its prevalence increases by 50% every decade. It is estimated that there may be an additional 100 million persons with asthma by 2025³.

According to WHO estimation 2,55,000 people died of asthma in 2005. Worldwide approximately 1,80,000 deaths are attributable to asthma every year. In Asia, increased prevalence is likely to be particularly dramatic in India and China³.

The total burden of asthma in India at an overall prevalence of 3% is estimated at over 30 million patients. In India, there is a prevalence of about 2.4% in adults over 15 year of age⁸.

In a study, asthma prevalence was calculated as Mumbai 3.5%, Delhi 2.28%, Kanpur 1.69% & Bangalore 3.47%⁶. In India, an estimated 57,000 deaths were attributable to asthma in 2004 and it was seen as one of the leading cause of mortality and morbidity in rural India⁴.

Globally the economic costs associated with asthma exceed those of tuberculosis and HIV/AIDS combines³. The cost of medication for asthma in India was estimated as US\$ 30 per month⁵.

Asthma is correlated with Eraippu in Siddha system of medicine. Takkolathi Chooranam” is a poly herbal Siddha medicine indicated for Eraippu which is mentioned in text “Agasthiyar 2000” 5th edition, October 2002 by Dr.S.Venkatarajan LIM and published by Saraswathi mahal noolagam¹⁵.

According to Siddha basic principle, karppu suvai cures inflammatory disease of the throat and removes sputum (phlegm) from the lungs, veppa verium and karppu pirivu pacifies kapham, and prapavam of karppu is to correct deranged kapham⁷.

The herbs in “Takkolathi chooranam” have karppu suvai, veppa verium and karppu pirivu and hence have great potential in treating Eraippu.

The above drug has not been evaluated so far for Eraippu, hence the author has selected “Takkolathi chooranam” to evaluate bronchodilator activity, anti-histaminic activity and its therapeutic efficacy.

AIM:

To evaluate the safety and efficacy of Takkolathi chooranam for Bronchodilator and Antihistaminic activity in the management of Eraippu (Bronchial asthma)

OBJECTIVE:**Primary objective:**

To evaluate the Bronchodilator and anti histaminic activity of Takkolathi chooranam for Eraippu (Bronchial asthma) in preclinical studies.

Secondary objective:

The efficacy of Takkolathi chooranam has been evaluated in the following aspect.

- Collection of literature evidences in Siddha aspect and Botanical aspect.
- Biochemical Analysis
- High Performance Thin Layer Chromatography
- Clinical study- a pilot study on trial medicine

STANDARD OPERATIVE PROCEDURE:

Collection and authentication of the raw drugs:

The raw drug were procured from raw drug store in Chennai and authenticated by competent authority of Department of Gunapadam, National Institute of Siddha, Chennai.

Ingredients:

Purified Takkolam	(Illicium verum)	- 1¼ varagan	(5.25 gms)
Purified Kirambhu	(Syzygium aromaticum)	- 1 ¼ varagan	(5.25 gms)
Purified Elakkai	(Elettaria cardamomum)	- 1¼ varagan	(5.25 gms)
Purified Thippili	(Piper longum)	- 2¼ varagan	(9.45 gms)
Purified Siruthekku	(Clerodendrum serratum)	- 5 varagan	(21 gms)
Purified Chukku	(Zingiber officinale)	- 10 varagan	(42 gms)
Purified Milagu	(Piper nigrum)	- 12½ varagan	(52.5 gms)
Sugar	(Saccharum officinarum)	- 33½ varagan	(140.7gms)

Purification process:

Purification of Takkolam¹³:

Takkolam was dried in the sunlight.

Purification of Kirambhu:

Kirambhu was dried in the sunlight¹³.

Purification of Elakkai:

Elakkai was dried in sunlight¹³.

Purification of Thippili:

Thippili was purified by soaking in the lemon juice¹⁴.

Purification of siruthekku:

Siruthekku was purified by removing the outer layer and cutted into small pieces, then they were dried in the sunlight¹³.

Purification of Chukku:

Chukku was purified by removing the outer layer and soaking in the limestone water¹⁴.

Purification of Milagu:

Milagu was soaked in buttermilk for 1 hour 15 minutes and then it was fried¹⁴.

Preparation of the medicine:

The raw drug was purified and pulverized by an electric grinder into fine powder, separately. And then it was sieved by using a fine silk cloth (vasthrakayam). The fine powder was purified by pittaviyal method. Then it was dried and ultra filtered by a cotton cloth and made into fine powder again. The powder was mixed with equal quantity of sugar and it was stored in a clean, dry air tight glass bottle¹⁵.

LABELLING:

Name of the preparation	:	Takkolathi chooranam
Taste	:	Kaarppu
Colour	:	Mild yellowish colour
Dose	:	6 gm b.d
Adjuvant	:	Luke warm water
Duration	:	1 month
Indication	:	Bronchial asthma
Date of manufacture	:	The drug was prepared in three batches. 13/4/12, 15/7/12, 16/10/12
Expiry	:	3 months from the date of manufacture

INGREDIENTS IN TAKKOLATHI CHOORANAM:

Illicium verum Hook.f



Syzygium aromaticum (Linn.)



Elettaria cardamomom (Linn.) Maton



Piper longum Linn



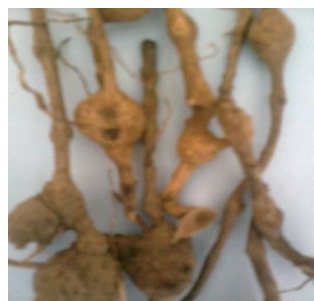
Zingiber officinale Rosc.



Piper nigrum Linn.



Clerodendrum serratum (Linn.)



Takkolathi chooranam



¾ Ì §, i Äö¹¹

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3. ÄðÄ · Ä ð çö¹⁷

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4. ¾ Ì Ç° ð¾ç Ý Ä½ö¹⁵

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5. ¾ Ì §, i Äî¾ç ÄîÆö²¹

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Research articles:

1. Research on antibacterial mechanism of essential oils and dominant monomer components of star anise²⁸
2. Study on chemical component analyzed by GC-MS and antioxidation of star anise extracts²⁹
3. Extraction and Free Radical Scavenging Investigation of Total Flavonones from Anise Leaves³⁰
4. Study on the antimicrobial stability of essential oil from star anise by supercritical carbondioxide extraction³¹
5. Chemical Composition and Antibacterial Activities of Illicium verum Against Antibiotic-Resistant Pathogens³²

§ÅÚ |ÅÄ÷, ù : « ÿ í ù, ÷ ù, ¼õ, ù ÖÅ;öì ù Å;öð, §°;°õ, ¾ÅÇ, ÅÄ; í ù

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Å;öð §°Öö þ · Åòð §Ç;öì ù ;É ÅÖóð ù:

1. « í Å;¾Ç ÝÄ½õ²⁰

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¾ Ò Ò \$ Ç ò : þ Ò Å ø, ò ò, ò ¾ ò, ò ý Å ò, Å Ò Á ý, ò Á ÷ Å
÷ ¾ ð Å Ò : « ÷ \$ ¾ Å ÷ ÷ Å ò ¾ Å ò ð Å Ò 1500

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$\frac{3}{4}i\tilde{n} \sim \frac{1}{4}, \frac{3}{4}i\tilde{u}, \tilde{A}_i\tilde{o}, \frac{1}{2}\tilde{u}\tilde{A}_i\tilde{o} \text{ p} \tilde{A}_i\tilde{C}\tilde{o} \sim \tilde{n} \frac{1}{4}i\tilde{l} \tilde{o} \tilde{S}_{\tilde{L}}i\tilde{o} \sim \tilde{C}\tilde{O}\tilde{o},$
 $\text{p}\tilde{O}\tilde{A}\tilde{o}, i\tilde{L}i \circ \tilde{y} \tilde{S}_{\tilde{L}}i \tilde{A}i \tilde{o}\tilde{I} \frac{3}{4}\tilde{O}\tilde{o}.$

$2\tilde{A}\tilde{o} \tilde{S}^{\tilde{O}}\tilde{O}\tilde{o} \text{ p} \tilde{A}\tilde{o}\tilde{O} \tilde{S}_{\tilde{L}}i\tilde{o}i \frac{1}{2}\tilde{E} \tilde{A}\tilde{O}\tilde{o}\tilde{D}_{\tilde{L}}\tilde{u}:$

1. $\frac{3}{4}\tilde{O}\tilde{o}\tilde{A}\tilde{C}\tilde{A}\tilde{C}\tilde{A}_i\frac{3}{4}\tilde{C}\tilde{I} \tilde{Y} \tilde{A}\frac{1}{2}\tilde{o}^{20}$

$\ll \tilde{C} \times \quad : \quad i\tilde{A}\tilde{O}\tilde{L}\tilde{E} \ll \tilde{C} \times, 2 \tilde{S}\tilde{A} \tilde{C}, 20i\tilde{u}$
 $\ll \tilde{U}\tilde{A}_i\tilde{E}\tilde{o} \quad : \quad \tilde{S}\frac{3}{4}\tilde{y}, i\tilde{L}\tilde{o}, \tilde{A}_i\tilde{D}\tilde{C}\tilde{o}\tilde{A}\tilde{E}i \circ i\tilde{U}$
 $\frac{3}{4}\tilde{O}\tilde{o} \tilde{S}_{\tilde{L}}i\tilde{o} \quad : \quad \text{p}\tilde{O}\tilde{A}\tilde{o}, \circ \tilde{C}, \circ \tilde{A}\tilde{o}, \circ \tilde{S}\tilde{A}\tilde{o}\tilde{I} \tilde{A}i \tilde{u}$
 $\neg \frac{3}{4}i\tilde{A}\tilde{o} \quad : \quad \ll \tilde{S}\frac{3}{4}\tilde{A} \div \tilde{A}\tilde{o}\frac{3}{4}\tilde{A} \frac{1}{2}\tilde{A}\tilde{A}\tilde{o} 1500$

2. $\tilde{A}\tilde{o}\tilde{A}_i\frac{3}{4}\tilde{C} \tilde{S}\tilde{A}_{\tilde{L}}\tilde{A}\tilde{o}^{20}$

$\ll \tilde{C} \times \quad : \quad i\tilde{L}i\tilde{O} \tilde{A}\tilde{o}\tilde{A}_i\tilde{l} \tilde{C} \times, 2 \tilde{S}\tilde{A} \tilde{C}, 1 \tilde{A}\tilde{n} \frac{1}{4}\tilde{A}\tilde{o}$
 $\frac{3}{4}\tilde{O}\tilde{o} \tilde{S}_{\tilde{L}}i\tilde{o} \quad : \quad \text{p}\tilde{O}\tilde{A}\tilde{o}, \circ \tilde{C}, i\tilde{y} \tilde{A}\tilde{o}, \tilde{S}\tilde{A}\tilde{u}\tilde{E}\tilde{o}, i\tilde{A}\tilde{A}_i\tilde{o} \frac{3}{4}$
 $\neg \frac{3}{4}i\tilde{A}\tilde{o} \quad : \quad \ll \tilde{S}\frac{3}{4}\tilde{A} \div \tilde{A}\tilde{o}\frac{3}{4}\tilde{A} \frac{1}{2}\tilde{A}\tilde{A}\tilde{o} 1500$

3. $\tilde{n} \frac{1}{4}i\tilde{o}\frac{3}{4}\tilde{C}\tilde{I} \tilde{S}\tilde{A}_{\tilde{L}}\tilde{A}\tilde{o}^{20}$

$\ll \tilde{C} \times \quad : \quad i\tilde{L}i\tilde{O} \tilde{A}\tilde{o}\tilde{A}_i\tilde{l} \tilde{C} \times, 2 \tilde{S}\tilde{A} \tilde{C}, 1 \tilde{A}\tilde{n} \frac{1}{4}\tilde{A}\tilde{o}$
 $\frac{3}{4}\tilde{O}\tilde{o} \tilde{S}_{\tilde{L}}i\tilde{o} \quad : \quad i\tilde{A}\tilde{o}\tilde{o}\tilde{I}, \tilde{o} \tilde{S}\tilde{A}_i\tilde{I} \text{ p}\tilde{O}\tilde{A}\tilde{o}, \tilde{A}_i\tilde{o}\frac{3}{4}\tilde{C}$
 $\neg \frac{3}{4}i\tilde{A}\tilde{o} \quad : \quad \ll \tilde{S}\frac{3}{4}\tilde{A} \div \tilde{A}\tilde{o}\frac{3}{4}\tilde{A} \frac{1}{2}\tilde{A}\tilde{A}\tilde{o} 1500$

4. $\tilde{u}\tilde{a}\tilde{A}_i\frac{3}{4}\tilde{C}\tilde{I} \tilde{Y} \tilde{A}\frac{1}{2}\tilde{o}^{23}$

$\ll \tilde{U}\tilde{A}_i\tilde{E}\tilde{o} \quad : \quad \tilde{S}\frac{3}{4}\tilde{y}$
 $\frac{3}{4}\tilde{O}\tilde{o} \tilde{S}_{\tilde{L}}i\tilde{o} \quad : \quad \text{p} \tilde{A}\tilde{o}\tilde{O}, \frac{1}{2}\tilde{o}\tilde{I} \tilde{A}_i\tilde{o}\tilde{o}, i\tilde{L}i \circ \tilde{A}\tilde{o}\tilde{O}$
 $\neg \frac{3}{4}i\tilde{A}\tilde{o} \quad : \quad \ll \tilde{S}\frac{3}{4}\tilde{A} \div \tilde{O}\frac{1}{4}\tilde{A} \tilde{A}_i\frac{1}{2}\tilde{o}$

$\frac{3}{4}\tilde{O}\tilde{o}\tilde{A}\tilde{C}\tilde{A}\tilde{C}\tilde{A}_i^{11}$

$\tilde{S}\tilde{A}\tilde{U}i\tilde{A}\tilde{A} \div \tilde{u} \quad : \quad \neg \div \frac{3}{4}\tilde{C}, \tilde{n} \circ \tilde{A}\tilde{o}, \tilde{A} \tilde{A}_i\tilde{o}\tilde{C}, \frac{1}{2}\tilde{A}\tilde{y}, i\tilde{S}\frac{1}{4}i\tilde{C}, \tilde{S}_{\tilde{L}}i\tilde{A}\tilde{o},$
 $\tilde{S}_{\tilde{L}}i\tilde{A}\tilde{C}, \circ \tilde{A}\tilde{o}, \circ i\tilde{E}, \tilde{D}\tilde{C}\tilde{A}\tilde{C}, \tilde{A}_i\frac{3}{4}\tilde{C}, \tilde{E}, \tilde{S}_{\tilde{L}}i \tilde{A}\tilde{A}\tilde{U}i \tilde{C}$

$\tilde{A}\tilde{A}\tilde{y}\tilde{A}\tilde{I} \tilde{o} \sim \tilde{U}\tilde{o}\tilde{O} \quad : \quad \tilde{L}i\tilde{o}, \ll i\tilde{C}\tilde{C}$

$i \tilde{A} \quad : \quad \tilde{L}i\tilde{o}\tilde{O}, \frac{3}{4}\tilde{y} \tilde{A}: i\tilde{A}\tilde{o}\tilde{A}\tilde{o}, \tilde{A}\tilde{C}\tilde{C} \times: \text{p}\tilde{E}\tilde{o}\tilde{O}$

$i \circ \tilde{o} \tilde{L} \quad : \quad i\tilde{A}\tilde{o}\tilde{A}\tilde{O}\tilde{n} \frac{1}{4}i\tilde{l} \tilde{C}, \ll \tilde{o}\tilde{I} \tilde{A}_i\tilde{o}\tilde{A}_{\tilde{L}}\tilde{u}\tilde{E}\tilde{C}$

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1. Immunomodulatory and antitumour activity of *Piper longum* Linn. and piperine³³
2. Histamine Release Inhibitory Activity of *Piper nigrum* Leaf³⁴

§ÅÚ |ÀÄ÷ : ÷ ñ Î ÄjÄí ÷

ÄÄýÄÎ õ - Úòð : þ ÷ Ä, §Å÷

Í ÷ Ä : ÷ ÷ òð, ÐÄ÷òð, ¾ý ÷ Ä: | ÄòÄò, Äç×: ÷ ÷ òð

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þ¾Éjø Óì ÷ ÷ ÷, þ ÷ ÄòÄÖÄø, Í Äò, ä ÷ ø Óýçç ÷ ÷ Ä, Äý
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°U\$¾ì §°Öð þ ÷ Äòð §çjðì ÷ jÉ ÄÖð ÷ ÷:

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2. °ç ÷ ¾ç Ý Ä ½õ¹⁸

¾Ö ÷ çç : Í Ä ÷ ÷ ÷, ÄÉ ÷, °Ä ÷
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3. þÄÄí ÷ ¾ç Ý Ä ½õ¹³

« Ç × : 1/2 §¾Äj, 2 §Ä ÷ Ç, 20çç
 ¾Ö ÷ çç : þ ÷ Äòð
 ÷ ¾Ä ÷ : °çç ÷ ÄòÉ ¾Ä ÷

4. $\frac{3}{4}i\ddot{A}\ddot{A}\acute{E}\ddot{A}\hat{i}\acute{Y}\ddot{A}\frac{1}{2}\ddot{o}$ ¹⁸

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$\neg\frac{3}{4}i\ddot{A}\ddot{o}$: « $\S_{\ddot{z}}\ddot{S}\frac{3}{4}\ddot{A}\ddot{\div}$ $\ddot{A}\ddot{o}\frac{3}{4}\ddot{A}\circ\frac{3}{4}i\ddot{A}\frac{1}{2}\ddot{t}$ | $\ddot{A}\ddot{n}\ddot{A}_i4000\pm\acute{y}\ddot{U}\ddot{o}\acute{A}\frac{1}{2}\ddot{t}4000$

5. $\acute{I}\ddot{n}\frac{1}{4}\ddot{A}_i\frac{3}{4}\ddot{t}\S\ddot{A}_{\ddot{z}}\ddot{A}\ddot{o}$ ²⁰

« $\S\times$: $\acute{I}_{\ddot{z}}i\ddot{O}''\frac{1}{4}\ddot{O}\ddot{A}_i\acute{I}_{\ddot{z}}\S\times$

$\frac{3}{4}\ddot{O}\ddot{o}\S_{\ddot{z}}i\ddot{o}$: $\S\ddot{A}\ddot{u}\ddot{E}\ddot{o}, \acute{A}\ddot{E}\circ\acute{I}_{\ddot{z}}\ddot{u}, p\ddot{O}\acute{A}\varnothing$

$\neg\frac{3}{4}i\ddot{A}\ddot{o}$: « $\S_{\ddot{z}}\ddot{S}\frac{3}{4}\ddot{A}\ddot{\div}$ $\ddot{A}\ddot{o}\frac{3}{4}\ddot{A}_{\ddot{z}}i\ddot{A}\ddot{A}\ddot{o}1500$

Research articles:

1. Antioxidant Effects of Roots of *Clerodendrum serratum* Linn³⁵

2. In-vitro and in-vivo antiasthmatic studies of *Clerodendrum serratum* Linn in Guinea pigs³⁶

Í Ì Î¹¹

$\S\ddot{A}\ddot{U}_{\ddot{z}}\ddot{A}\ddot{A}\ddot{\div}_{\ddot{z}}\ddot{u}$: « $\ddot{O}\acute{I}_{\ddot{z}}\acute{y}, \ll\frac{3}{4}_{\ddot{z}}\ddot{o}, \neg\div\ddot{O}\ddot{A}_{\ddot{z}}\ddot{o}, \neg\acute{A}\ddot{I}\varnothing\ddot{A}\ddot{o}, \acute{I}\ddot{A}\ddot{o}\frac{3}{4}\ddot{A}\ddot{o},$
 $\acute{I}\ddot{n}\ddot{E}\acute{I}_{\ddot{z}}i\ddot{n}\ddot{E}, \acute{I}^{\circ a}\ddot{A}\acute{y}\ddot{E}\ddot{o}, \S\ddot{A}\acute{I}\ddot{U}, \S_{\ddot{z}}i\ddot{A}\ddot{o}, \acute{A}\ddot{A}\ddot{a}\ddot{E}\ddot{A}$
 $\ll\acute{A}\ddot{t}\frac{3}{4}\ddot{o}$

$\ddot{A}\ddot{A}\acute{y}\ddot{A}\hat{i}\ddot{o}\neg\acute{U}\ddot{o}\ddot{d}$: $\S_{\ddot{z}}\ddot{A}\acute{I}\ddot{I}(\neg\ddot{A}\div\ddot{O}\frac{3}{4}\ddot{D})$

$\acute{I}''\ddot{A}$: $\S_{\ddot{z}}i\div\ddot{O}\ddot{d}, \frac{3}{4}\acute{y}''\acute{A}:\ddot{A}\ddot{o}\ddot{A}\ddot{o}, \acute{A}\ddot{t}\times:\S_{\ddot{z}}i\div\ddot{O}\ddot{d}$

$\acute{I}^{\circ}\ddot{o}''_{\ddot{z}}$: $\acute{A}\ddot{o}\frac{3}{4}\ddot{D}\ddot{a}\ddot{n}\ddot{E}, \ddot{I}\ddot{A}\ddot{o}\ddot{A}\ddot{O}\ddot{n}\frac{1}{4}i\ddot{I}_{\ddot{z}}\ddot{t}, \ll\S_{\ddot{z}}\ddot{O}\hat{i}\ddot{A}_i\ddot{o}\ddot{A}_{\ddot{z}}\ddot{u}\ddot{E}\ddot{t}$

$\ddot{I}\ddot{A}_i\ddot{D}\ddot{I}\frac{1}{2}\ddot{o}$: $\acute{Y}''\ddot{A}\ddot{A}\ddot{o}\frac{3}{4}\ddot{o}\acute{I}_{\ddot{z}}\acute{I}^{\circ}i\ddot{t}\ddot{O}\ddot{d}\S\frac{3}{4}i\frac{1}{4}\S\ddot{A}\ddot{o}\ddot{A}\ddot{o}\acute{A}\ddot{E}''\ddot{A}$
 $\ddot{a}\ddot{A}\ddot{o}p''\ddot{A}\ddot{o}\ddot{A}\ddot{O}\ddot{A}\ddot{o}\ddot{a}\acute{I}\ddot{I}\S_{\ddot{z}}\neg\ddot{A}_i\ddot{A}_{\ddot{z}}\ddot{A}$
 $\S\frac{3}{4}i\frac{1}{4}\ddot{A}\frac{3}{4}\ddot{t}\circ\ddot{I}\ddot{A}\ddot{o}\acute{I}_{\ddot{z}}i\frac{1}{4}\ddot{A}_i\frac{3}{4}\acute{I}\acute{y}\acute{A}\ddot{z}\ddot{E}\ddot{o}$
 $\S\frac{3}{4}i\frac{1}{4}\ddot{o}\neg\ddot{A}\ddot{o}\S\ddot{A}_i\ddot{I}\ddot{I}\acute{I}\acute{I}\ddot{I}.$

$\acute{I}\ddot{I}_{\ddot{z}}\ddot{t}\acute{E}_{\ddot{z}}i\varnothing p''\ddot{A}\ddot{o}\ddot{d}, p\ddot{O}\acute{A}\varnothing, \S\ddot{A}\ddot{u}\ddot{E}\ddot{o}, \S_{\ddot{z}}i\ddot{D}\ddot{I}\ddot{I}\ddot{o}\frac{3}{4}\varnothing, \acute{O}_{\ddot{z}}\S_{\ddot{z}}i\ddot{o}, \frac{3}{4}''\ddot{A}$
 $\S_{\ddot{z}}i\ddot{o}, 3\ddot{A}\acute{I}\ddot{A}\ddot{o}\S\ddot{A}_i\ddot{o}.$

Í Ì Î $\S^{\circ}\ddot{O}\ddot{o}p''\ddot{A}\ddot{o}\ddot{d}\S_{\ddot{z}}i\ddot{o}\acute{I}_{\ddot{z}}i\acute{E}\acute{A}\ddot{O}\ddot{o}\ddot{D}_{\ddot{z}}\ddot{u}$:

1. $\circ\acute{A}\acute{y}\acute{Y}\ddot{A}\frac{1}{2}\ddot{o}$ ¹⁸

$\frac{3}{4}\ddot{O}\ddot{o}\S_{\ddot{z}}i\ddot{o}$: $\acute{I}\ddot{A}_i\circ\ddot{o}, \S_{\ddot{z}}i\circ\ddot{o}$

$\neg\frac{3}{4}i\ddot{A}\ddot{o}$: « $\S_{\ddot{z}}\ddot{S}\frac{3}{4}\ddot{A}\ddot{\div}$ $\ddot{A}\ddot{o}\frac{3}{4}\ddot{A}\circ\frac{3}{4}i\ddot{A}\frac{1}{2}\ddot{t}$ | $\ddot{A}\ddot{n}\ddot{A}_i4000\pm\acute{y}\ddot{U}\ddot{o}\acute{A}\frac{1}{2}\ddot{t}4000$

2. $\hat{a}_{\hat{z}_i} \hat{Y} \tilde{A}^{1/2} \tilde{o}^{13}$

$\frac{3}{4} \tilde{O} \tilde{o} \S_{\hat{z}_i} \tilde{o} : \quad {}^\circ \tilde{A} \tilde{o}, \hat{z}_i {}^\circ \tilde{o}$

$\neg \frac{3}{4} \hat{z}_i \tilde{A} \tilde{o} : \quad {}^\circ \tilde{c}_i \tilde{o} \hat{z}_i \tilde{A} \tilde{o} \tilde{E} \frac{3}{4} \tilde{A} \tilde{o}$

3. $\hat{A}_i \div \tilde{A} \tilde{o} \frac{3}{4} \tilde{A}_i \frac{3}{4} \tilde{c} \hat{Y} \tilde{A}^{1/2} \tilde{o}^{18}$

$\frac{3}{4} \tilde{O} \tilde{o} \S_{\hat{z}_i} \tilde{o} : \quad \hat{I} \hat{A}_i {}^\circ \hat{z}_i {}^\circ \tilde{o}$

$\neg \frac{3}{4} \hat{z}_i \tilde{A} \tilde{o} : \quad \ll \hat{z}_i \tilde{S} \frac{3}{4} \tilde{A} \div \hat{z}_i \tilde{A} \tilde{o} \frac{3}{4} \tilde{A} {}^\circ \tilde{o} \frac{3}{4} \hat{z}_i \tilde{A}^{1/2} \hat{z}_i \hat{A} \tilde{n} \hat{A}_i 4000 \pm \hat{y} \hat{U} \tilde{o} \hat{A}^{1/2} \hat{z}_i 4000$

$4 {}^\circ \tilde{c} \hat{A} \hat{z}_i \frac{3}{4} \hat{I} \hat{Y} \tilde{A}^{1/2} \tilde{o}^{20}$

$\ll \hat{C} \times : \quad \hat{z}_i \tilde{A} \tilde{O} \hat{z}_i \tilde{E} \ll \hat{C} \times, 2 \S \hat{A} \hat{z}_i \hat{C}$

$\ll \hat{U} \hat{A}_i \tilde{E} \tilde{o} : \quad \S \frac{3}{4} \hat{y}, \hat{p} \hat{z}_i {}^\circ \tilde{c} \hat{z}_i \hat{U}$

$\frac{3}{4} \tilde{O} \tilde{o} \S_{\hat{z}_i} \tilde{o} : \quad \hat{p} \tilde{O} \hat{A} \tilde{o}, {}^\circ \hat{z}_i \hat{C}$

$\neg \frac{3}{4} \hat{z}_i \tilde{A} \tilde{o} : \quad \ll \hat{z}_i \tilde{S} \frac{3}{4} \tilde{A} \div \hat{z}_i \tilde{A} \tilde{o} \frac{3}{4} \tilde{A} \hat{z}_i \hat{A} \tilde{A} \tilde{o} 1500$

$\hat{A} \hat{C} \hat{I}^{11}$

$\S \hat{A} \hat{U} \hat{z}_i \hat{A} \hat{A} \div \hat{z}_i \hat{U} : \quad \hat{z}_i \tilde{A} \hat{z}_i \tilde{E}, \hat{z}_i \tilde{E} \hat{z}_i \tilde{A} \tilde{o}, \S_{\hat{z}_i} \hat{C} \hat{z}_i \tilde{o}, \frac{3}{4} \tilde{c} \hat{I} \hat{z}_i \hat{U}, \hat{A} \hat{z}_i \tilde{A} \tilde{o},$
 ${}^\circ \tilde{O} \hat{A} \tilde{A} \tilde{o} \frac{3}{4} \tilde{o}, \hat{A} \hat{U} \hat{C} {}^\circ \tilde{o}, \hat{A}_i {}^\circ \tilde{o}, \hat{A} \hat{z}_i \hat{A} \hat{A}_i \hat{C} \hat{z}_i$

$\hat{A} \hat{A} \hat{y} \hat{A} \hat{I} \tilde{o} \hat{z}_i \hat{U} \tilde{o} : \quad \hat{A} \hat{z}_i \frac{3}{4}, \hat{z}_i \hat{z}_i \hat{E}$

$\hat{I} \hat{z}_i \hat{A} : \quad \hat{z}_i \hat{z}_i \tilde{o} \tilde{o}, \hat{z}_i \div \tilde{o} \tilde{o}, \frac{3}{4} \hat{y} \hat{z}_i \hat{A} : \hat{A} \tilde{o} \tilde{A} \tilde{o}, \hat{A} \hat{z}_i \times : \hat{z}_i \div \tilde{o} \tilde{o}$

$\hat{z}_i {}^\circ \tilde{o} \hat{z}_i : \quad \hat{z}_i \tilde{E} \tilde{O} \tilde{n} \frac{1}{4} \hat{z}_i \tilde{c}, \hat{z}_i \tilde{A} \tilde{o} \tilde{A} \tilde{O} \tilde{n} \frac{1}{4} \hat{z}_i \tilde{c}, \ll \hat{z}_i \tilde{o} \hat{I} \hat{A}_i \tilde{o} \hat{A} \hat{z}_i \tilde{u} \tilde{E} \hat{z}_i,$
 $\hat{O} \hat{z}_i \tilde{E} \hat{z}_i \tilde{A} \tilde{o} \hat{A} \hat{z}_i \tilde{u} \tilde{E} \hat{z}_i, \frac{3}{4} \tilde{E} \tilde{o} \tilde{o} \tilde{n} \frac{1}{4} \hat{z}_i \tilde{c}, \hat{A} \hat{z}_i \hat{z}_i \hat{z}_i \hat{A} \hat{z}_i {}^\circ \tilde{c},$
 $\hat{A}_i \frac{3}{4} \hat{A} \frac{1}{4} \hat{z}_i \tilde{c}, \hat{z}_i \hat{I} {}^\circ \hat{z}_i \hat{c}$

$\hat{z}_i \hat{A}_i \tilde{D} \hat{I}^{1/2} \tilde{o} : \quad$

${}^\circ \tilde{B} \hat{I} \tilde{A} \tilde{o} \hat{A}_i \tilde{n} \hat{I} {}^\circ \tilde{S} \tilde{A} \tilde{o} \hat{A} \hat{I} \hat{z}_i \tilde{A}_i^{1/2} \hat{z}_i \hat{y} \hat{A} \tilde{o}$

$\hat{A}_i \frac{3}{4} \tilde{o} \ll \tilde{O} {}^\circ \tilde{A} \tilde{o} \frac{3}{4} \tilde{o} \hat{A}_i \tilde{a} \tilde{A} \tilde{o}$ - ஒதுசன்னி

$\hat{A}_i {}^\circ \hat{A} \tilde{S} \hat{A}_i \tilde{A} \tilde{o} \ll \frac{1}{4} \hat{y} \S \hat{A} \hat{z}_i \tilde{o} \hat{z}_i {}^\circ \hat{A} \hat{z}_i \hat{A}$

$\hat{z}_i {}^\circ \hat{I} \hat{z}_i \tilde{E} \hat{A} \hat{C} \hat{z}_i \tilde{E} \hat{z}_i \tilde{o}$

$\hat{p} \frac{3}{4} \tilde{E} \hat{z}_i \tilde{o} \hat{z}_i \hat{C} \hat{z}_i \hat{I} \tilde{A} \tilde{o}, \hat{A}_i \tilde{n} \hat{I}, \S_{\hat{z}_i} \hat{z}_i \hat{E}, \hat{p} \tilde{O} \hat{A} \tilde{o}, \hat{z}_i {}^\circ \hat{A} \hat{A} \tilde{A} \hat{z}_i, \hat{p} \tilde{A} \tilde{o} \frac{3}{4} \hat{z}_i \hat{y} \hat{A} \tilde{o},$
 $\hat{z}_i \hat{A}_i \hat{z}_i \hat{A} \hat{p} \hat{z}_i \hat{A} \S \hat{A}_i \hat{I} \tilde{o}.$

ÁŒĬ Œ°Œō þ̄̄ Åðð ŒĴiöĬ ŒĴÉ ÅŒōĐ Œ:

1. Í ÅĬ°Ĭ Œ¼ĬĬ Œ̄ Å̄½ō²⁰

« ŪÅĬÉō : Œ¼Œ, þ̄̄ °Œ°ĬŪ, ĬĴō

¾Œō ŒĴiö : þŒÅø, Í ÅĬ°ŒĴiö Œ̄ Œ̄i°ō, ®̄̄ Ç

→ ¾ĬÅō : « Œ̄ Š¾Å÷̄̄ Åð¾Å ŒĬÅō 1500

2. ÅĬÅĬ¼Ĭ Œ̄ Å̄½ō²⁰

« Ç× : ĬÅō Œ̄ « Ç×, 2 ŒÅ̄̄ Ç, 1 Åñ ¼Åō

« ŪÅĬÉō : Œ¼Œ, þ̄̄ °Œ°ĬŪ, ĬĴō

¾Œō ŒĴiö : þŒÅø, ÅĬó¾Ĵ, ĬÅð̄̄ ¼

→ ¾ĬÅō : « Œ̄ Š¾Å÷̄̄ Åð¾Å ŒĬÅō 1500

3. þÅ°Ĭ Ĭ°óà Åō²²

« Ç× : Å½ Ĭ̄ Å̄̄ ¼, 2 ŒÅ̄̄ Ç, 7 ĴiŪ

« ŪÅĬÉō : °Å Œ̄ ÅĬÉō

¾Œō ŒĴiö : Œ̄i°ō, °Åō, þŒÅø, þ̄̄ Çðð

→ ¾ĬÅō : Œ̄ Å̄̄ Å̄÷̄̄ Åð¾Å 1000

4. Œ̄ñ ¼Ĭ Œ̄¾ĴĬ Œ̄ Å̄½ō²³

« Ç× : ÅĬ Ĭ̄ Ç×

¾Œō ŒĴiö : þŒÅø, Í ÅĬ°Œ̄i°ō

→ ¾ĬÅō : « Œ̄ Š¾Å÷̄̄ « ð¼Å̄̄ ½ ÅĬ¼ō

Research articles:

1. The antioxidant and radical scavenging activities of black pepper (*Piper nigrum*) seeds³⁷
2. Efficacy of piperine, an alkaloidal constituent from *Piper nigrum* on erythrocyte antioxidant status³⁸
3. Anti-inflammatory activity of piperine³⁹.

TAKKOLAM

BOTANICAL NAME²⁴ : Illicium verum Hook.f

CLASSIFICATION²⁵:

Kingdom	:	Plantae– Plants
Subkingdom	:	Tracheobionta– Vascular plants
Superdivision	:	Spermatophyta– Seed plants
Division	:	Magnoliophyta– Flowering plants
Class	:	Magnoliopsida– Dicotyledons
Subclass	:	Magnoliidae
Order	:	Illiciales
Family	:	Illiciaceae– Star-anise family
Genus	:	Illicium L.– anisetree
Species	:	I.verum Hook. f. – staranise tree

VERNACULAR NAMES: Star anise, star aniseed, or Chinese star anise

BOTANICAL DESCRIPTION²⁶: An annual herb about 0.5m high; root fusiform, and the stem is erect, round, leaves are petiolate, inflorescences are medium sized umbels, and fruit is ovate to oblong and flattened at the sides.

DISTRIBUTION: A medium-sized native evergreen tree of northeast Vietnam ,southwest China.

PARTS USED: Fruit.

CHEMICAL CONSTITUENTS: Anethole, cinnamaldehyde, 0-methoxycinnamaldehyde, p-methoxy-cinnamaldehyde, cinnamic acid, p-methoxycinnamaldehyde, cinnamyl alcohol, trans-anethole, beta-caryophyllene, citronellol, estragole, eugenol methyl ether, myrcene.

PHARMACOLOGICAL ACTIVITY: Antibacterial, Appetizer, Carminative, Expectorant, Stimulant

ADULTERANTS: Japanese star anise (Illicium anisatum) is adulterated with star anise.

KIRAMBHU

BOTANICAL NAME²⁴: *Syzygium aromaticum* (Linn.) Merrill & Perry.

CLASSIFICATION²⁵:

Kingdom	:	Plantae– Plants
Subkingdom	:	Tracheobionta– Vascular plants
Superdivision	:	Spermatophyta– Seed plants
Division	:	Magnoliophyta– Flowering plants
Class	:	Magnoliopsida– Dicotyledons
Subclass	:	Rosidae
Order	:	Myrtales
Family	:	Myrtaceae– Myrtle family
Genus	:	<i>Syzygium</i> P. Br. ex Gaertn.– syzygium
Species	:	<i>S.aromaticum</i> (L.) Merr. & L.M. Perry– clove

VERNACULAR NAMES:

- **Eng.-** Clove tree,Cloves
- **Hindi-** Lavanga
- **Kan.-**Lavanga
- **Mal.-**Karampu
- **Tam.-**Kirambu, Lavangam
- **Tel.-**Lavangalu

BOTANICAL DESCRIPTION: A pyramidal or conical evergreen upto 12m high leaves simple, flower buds greenish to pink, fruits fleshy, dark pink drupes, seeds grooved on one side, obolong.

DISTRIBUTION: It is a native of islands of Malay Archipelago, especially Moluccas. In India, it is reported to be grown in Tamil Nadu (Nilgris, Courtallam and Kanyakumari) and Kerala.

PARTS USED: Flower buds, oil.

ACTIONS: Digestive, carminative, stomachic, stimulant, aphrodisiac, appetizer, expectorant, emollient, rejuvenating and tonic.

Physical constants: Foreign matter- Not more than 2%; Total ash- Not more than 7%; Acid-insoluble ash- Not more than 1%; Alcohol-soluble extractive- Not less than 3%; Water-soluble extractive- Not less than 9%; Volatile oil- Not less than 15%.

CHEMICAL CONSTITUENTS: Polyoxygenated chromone C-glucoside, isobiflorin, eugenol, acetyleugenol, epoxydihydrocaryophyllene, caryophellene and furfural traces, naphthalene, ellagitannin- eugeniin, caryophyllene oxide.

PHARMACOLOGICAL ACTIVITY: Anticarcinogenic, antioxidant, antibacterial, antiviral, radical scavenging activity, histamine release inhibitory activity.

ADULTERANTS: Cloves are adulterated with unripe fruits of *Cinnamomum verum*.

ELLAM

BOTANICAL NAME²⁴: *Elettaria cardamomom* (Linn.) Maton.

CLASSIFICATION²⁵:

Kingdom	:	Plantae– Plants
Subkingdom	:	Tracheobionta– Vascular plants
Superdivision	:	Spermatophyta– Seed plants
Division	:	Magnoliophyta– Flowering plants
Class	:	Liliopsida– Monocotyledons
Subclass	:	Zingiberidae
Order	:	Zingiberales
Family	:	Zingiberaceae– Ginger family
Genus	:	<i>Elettaria</i> Maton– <i>elettaria</i>
Species	:	<i>E.cardamomum</i> (L.) Maton– cardamom

VERNACULAR NAMES:

- **Eng.-** Cardamom
- **Hindi-** choti ilayachi
- **Kan.-**Elakki
- **Mal.-**Elam

- **Tam.-**Ellakay
- **Tel.-**Chinne elakulu

BOTANICAL DESCRIPTION: A tall herbaceous perennial, leaves 30-90cm long, flowers white or pale green with a central lip straked with violet, fruits trilocular, seeds 15-20 per pod.

DISTRIBUTION: It is a native of the moist evergreen forests of South India, growing wild in the Western Ghats, between 800-1600 m. It is commonly cultivated in Kerala, Karnataka and Tamilnadu.

PARTS USED: Seed

ACTIONS: Seeds are aromatic, cooling, stimulant, carminative, digestive, stomachic, diuretic, carditonic, alexiteric, expectorant, tonic and abortifacient.

Physical constants: Total ash- Not more than 6%; Acid- insoluble ash- Not more than 4%; Alcohol- soluble extractive- Not less than 2%; Water-soluble extractive- Not less than 10%; Volatile oil- Not less than 4%.

CHEMICAL CONSTITUENTS: α -Pinene, sabinene, myrcene, limonene, cineol, cymene, methyl heptenone, linalool, linalyl acetate, α & β -terpeneol, α -terpinyl acetate, borneol, neryl acetate, geraniol, nerol, nerolidol, heptacosane, 1,8-cineole, camphene, terpinene, α -humelene.

PHARMACOLOGICAL ACTIVITY: Hepatoprotective, anti-inflammatory, analgesic, antispasmodic, antimicrobial, antifugal.

SUBSTITUTES: Amomum subulatum Roxb., possesses similar properties to that of Elettaria cardamomum for which they are often substituted.

THIPPILI

BOTANICAL NAME²⁴ : Piper longum Linn

CLASSIFICATION²⁵:

Kingdom	:	Plantae– Plants
Subkingdom	:	Tracheobionta– Vascular plants

Superdivision	:	Spermatophyta– Seed plants
Division	:	Magnoliophyta– Flowering plants
Class	:	Magnoliopsida– Dicotyledons
Subclass	:	Magnoliidae
Order	:	Piperales
Family	:	Piperaceae– Pepper family
Genus	:	Piper L.– pepper
Species	:	<i>P. longum</i> L.– Indian long pepper

VERNACULAR NAMES

- **Eng.-**Indian long pepper
- **Hindi-**Pipli
- **Kan.-**Hipli
- **Tam.-**Tippili
- **Tel.-**Pipallu

BOTANICAL DESCRIPTION: A slender, aromatic climber with perennial woody roots, stems creeping, leaves ovate, spikes cylindrical, pedunculate, fruits ovoid, yellowish orange.

DISTRIBUTION: It occurs in hotter parts of India from Central Himalayas to Assam, evergreen forests of Western Ghats from Konkan to Travancore

PARTS USED: Fruit, root

ACTIONS: Tonic, diuretic, purgative, expectorant, anthelmintic, stomachic, digestive and emmenagogue.

CHEMICAL CONSTITUENTS: Terpinolene, zingiberene, p-cymene, piperine, pipartine, triacontane, dihydro-stigmasterol, piperine and sesamin, caryophyllene, a sesquiterpene alcohol.

PHARMACOLOGICAL ACTIVITY: Antibacterial, anti-inflammatory, antitubercular, anthelmintic, hypoglycaemic, hepatoprotective.

SIRUTHEKKU

BOTANICAL NAME²⁴ : *Clerodendrum serratum* (Linn.) Moon

CLASSIFICATION²⁶:

Kingdom	:	Plantae
Phylum	:	Eudicota
Class	:	Angiospermae
Subclass	:	Lamiidae
Order	:	Lamiales
Family	:	Verbenaceae
Genus	:	<i>Clerodendrum</i>
Species	:	<i>C.serratum</i>

VERNACULAR NAMES

- **Hindi**-Bharangi
- **Kan.**-Gantabarangi
- **Mal.**- Cerutekku
- **Tam.**-Sirutekku
- **Tel.**-Barangi

BOTANICAL DESCRIPTION: Perennial herbs or shrubs, leaves sessile or nearly so, flowers blue-purple or white, drupes 6 mm long, broadly obovoid, rather succulent, dark-purple when ripe.

DISTRIBUTION: More or less throughout India, in forests upto 1500 m altitude.

PARTS USED: Root, leaf.

ACTIONS: Antiinflammatory, digestive, carminative, stomachic, stimulant, expectorant and febrifuge.

CHEMICAL CONSTITUENTS: Sterratagenic acid, queretaroic acid, some phytosterols, saponins, ferulic acid, arabinose, scutellarein, baicalein.

PHARMACOLOGICAL ACTIVITY: Antihistaminic, hypotensive, bronchoconstrictor, antiallergic, antiasthmatic, antibiotic and stomachic.

SUBSTITUTES AND ADULTERANTS: The bark of *Gardenia turgida* Roxb. is an adulterated to be sold as Bharangi bark. Substituted by root of *Solanum surattense* Burm.f.

CHUKKU

BOTANICAL NAME²⁴ : *Zingiber officinale* Rosc.

CLASSIFICATION²⁵:

Kingdom	:	Plantae– Plants
Subkingdom	:	Tracheobionta– Vascular plants
Superdivision	:	Spermatophyta– Seed plants
Division	:	Magnoliophyta– Flowering plants
Class	:	Liliopsida– Monocotyledons
Subclass	:	Zingiberidae
Order	:	Zingiberales
Family	:	Zingiberaceae– Ginger family
Genus	:	<i>Zingiber</i> Mill.– ginger
Species	:	<i>Z.officinale</i> Roscoe– garden ginger

VERNACULAR NAMES

- **Eng.-**Ginger
- **Hindi-**Sonth
- **Kan.-**Shunthi
- **Tam.-**Chukku
- **Tel.-**Sonthi

BOTANICAL DESCRIPTION: Perennial herb; leaves sessile, flowers greenish with a small acute , sheath 10-15 cm long, fruits oblong capsules.

DISTRIBUTION: It is mostly cultivated in Kerala and run wild in some places in Western Ghats.

PARTS USED: Fresh rhizome and dried rhizome

ACTIONS: The raw ginger is acrid, thermogenic, carminative, laxative and digestive.

Physical constants: Dried rhizome: - Total ash- Not more than 6%; Water soluble ash- Not less than 1.5%; Alcohol (90%) soluble extractive- Not less than 3%; Water soluble extractive- Not less than 10%.

CHEMICAL CONSTITUENTS: Heptane, octane, isovaleraldehyde, nonanol, ethyl pinene, camphene, pinene, sabinene, myrecene, limonene, sesquiphellandrene, gingerol, zingerone, shogaol, dihydrogingerol.

PHARMACOLOGICAL ACTIVITY: Antiinflammatory, hypolipidaemic, antiemetic, antiulcer, antioxidant, antibacterial, hepatoprotective, hypoglycaemic.

SUBSTITUTES AND ADULTERANTS: The rhizomes of *Z. casummar* Roxb. are sometimes used as substitute to *Z. officinale*.

MILAGU

BOTANICAL NAME²⁴ : *Piper nigrum* Linn.

CLASSIFICATION²⁵:

Kingdom	:	Plantae– Plants
Subkingdom	:	Tracheobionta– Vascular plants
Superdivision	:	Spermatophyta– Seed plants
Division	:	Magnoliophyta– Flowering plants
Class	:	Magnoliopsida– Dicotyledons
Subclass	:	Magnoliidae
Order	:	Piperales
Family	:	Piperaceae– Pepper family
Genus	:	<i>Piper</i> L.– pepper
Species	:	<i>P. nigrum</i> L.– black pepper

VERNACULAR NAMES

- **Eng.-**Black pepper
- **Hindi-**Kalimirsch
- **Kan.-**Kaalumenasu
- **Tam.-**Milagu
- **Tel.-**Miriyalu

BOTANICAL DESCRIPTION: Much branched climbing shrub, leaves simple, alternate, cordate, flowers minute, fruits ovoid or globose, seeds globose, perisperm hard and white.

DISTRIBUTION: It is mostly found cultivated in the hot and moist parts of India, Srilanka and other tropical countries. The plant is wild in Western Ghats and South India at 500 – 1600 m in evergreen forests.

PARTS USED: Fruit

ACTIONS: Carminative, digestive, stimulant and stomachic.

Physical constants: Total ash- Not more than 5.0%; Acid insoluble ash- Not more than 0.5%; Alcohol soluble extractive- Not less than 6.0%; Water soluble extractive- Not less than 6.0%.

CHEMICAL CONSTITUENTS: Pipernol, piperine, piperoleine A& B, pellitonin, methylenedioxycinnamic acid, piperettine, ascorbic acid, carotene, sitosterol.

PHARMACOLOGICAL ACTIVITY: Antioxidant, antipyretic, anti-inflammatory, antibacterial.

ADULTERANTS: Carica papaya Linn. Seeds are used to adulterate black pepper.

PHYSICAL PROPERTIES OF TAKKOLATHI CHOORANAM

The physical properties of Takkolathi Chooranam were carried at Sri Ramachandra University, Chennai.

pH at 10% of aqueous solution:

Five grams of Takkolathi Chooranam was weighed accurately and placed in clear 100 ml beaker. Then 50 ml of distilled water was added to it and dissolved well. After 30 minutes it was then applied in to pH meter at standard buffer solution of 4.0,7.0,9.2.

Ash Values

The Ash values are a measure of the inorganic constituents present in the raw drug. A high ash content explains its unsuitable nature to be used as a drug

Total Ash

A little of extract was taken in a silica crucible previously ignited, cooled and weighed. It was incinerated by gradually increasing the heat not exceeding dull red heat (450°C) until free from carbon, cooled and weighed. The percentage of ash was calculated with reference to air- dried drug. The procedure was repeated to get the constant weight.

Water soluble ash

The total ash was boiled with 25 ml water and filtered through ash less filter paper (Whatmann 4.1). It was followed by washing with hot water .The filter paper was dried and ignited in the silica crucible, cooled and the water insoluble ash was weighed. The water-soluble ash can be calculated by subtracting the water insoluble ash from the total ash.

Acid insoluble ash

The total ash obtained was boiled for 5 minutes with 25 ml of (10% w/v) dilute hydrochloric acid and filtering through ash less filter paper (Whatmann 4.1). The filter paper was ignited in the silica crucible, cooled and insoluble ash was weighed. (Table A)

BIO -CHEMICAL ANALYSIS OF TAKKOLATHI CHOORANAM -

The biochemical analysis of the Takkolathi Chooranam was carried out in the Biochemistry lab, NIS

S.No	EXPERIMENT	OBSERVATION	INFERENCE
1	Appearance of sample	Light yellow in colour	
2	Solubility: a. A little(500mg) of the sample was shaken well with distilled water. b. A little(500mg) of the sample was shaken well with con. HCl/Con. H ₂ SO ₄	Sparingly soluble	Absence of Silicate
3	Action of Heat: A small amount(500mg) of the sample was taken in a dry test tube and heated gartly at first and then strong.	No white fumes evolved	Absence of Carbonate
4	Flame Test: A small amount(500mg) of the sample was made into a paste with con. HCl in a watch glass and introduced into non-luminous part of the Bunsen flame.	No Bluish green flame appeared.	Absence of Copper
5	Ash Test: A filter paper was soaked into a mixture of sample and dil. cobalt nitrate solution and introduced into the Bunsen flame and ignited.	No yellow colour flame appeared.	Absence of sodium

Preparation of Extract:

5gm of Takkolathi Choornam was weighed accurately and placed in a 250ml clean beaker and added with 50ml of distilled water. Then it was boiled well for about 10 minutes. Then it was cooled and filtered in a 100ml volumetric flask and made up to 100ml with distilled water.

S.No	EXPERIMENT	OBSERVATION	INFERENCE
	I. Test For Acid Radicals		
1.	Test For Sulphate: a. 2ml of the above prepared extract was taken in a test tube to this added 2ml of 4% dil ammonium oxalate solution b. 2ml of the above prepared extracts was added with 2ml of dil-HCl was added until the effervescence ceases off. Then 2ml of dil. Barium chloride solution was added.	Cloudy appearance present	Presence of Sulphate
2.	Test For Chloride: 2ml of the above prepared extract was added with dil. HCl till the effervescence ceases. Then 2ml of dil. silver nitrate solution was added.	No cloudy appearance.	Absence of Chloride
3.	Test For Phosphate: 2ml of the extract was treated with 2ml of dil. ammonium molybdate solution and 2ml of con. HNO ₃ .	No Yellow appearance present	Absence of Phosphate
4	Test For Carbonate: 2ml of the extract was treated with 2ml dil. Magnesium sulphate solution	No Cloudy appearance.	Absence of carbonate
5.	Test For Nitrate: 1gm of the substance was heated with copper turning and concentrated H ₂ SO ₄ and viewed the test tube vertically down.	No Brown gas evolved.	Absence of Nitrate
6.	Test For Sulphide: 1gm of the substance was treated with 2ml of con. HCL	No Rotten Egg Smelling gas.	Absence of Sulphide
7.	Test For Fluoride & Oxalate: 2ml of extract was added with 2ml of dil. Acetic acid and 2ml dil. calcium chloride solution and heated.	No Cloudy appearance	Absence of fluoride and oxalate

8.	Test For Nitrite: 3drops of the extract was placed on a filter paper, on that-2 drops of dil.acetic acid and 2 drops of dil.Benzidine solution was placed.	No Characteristic changes	Absence of Nitrite
9.	Test For Borate: 2 Pinches(50mg) of the substance was made into paste by using dil.sulphuric acid and alcohol (95%) and introduced into the blue flame.	No Bluish green colour flame.	Absence of borate
II. Test For Basic Radicals			
1.	Test For Lead: 2ml of the extract was added with 2ml of dil.potassium iodine solution.	No yellow Precipitate obtained.	Absence of Lead
2.	Test For Copper: a. One pinch(50mg) of substance was made into paste with con. HCl in a watch glass and introduced into the non-luminous part of the flame.	No Blue colour flame No Blue colour precipitate formed.	Absence of copper
3.	Test For Aluminium: To the 2ml of extract, dil.sodium hydroxide was added in 5 drops to excess.	No Yellow colour appeared.	Absence of aluminium
4.	Test For Iron: a. To the 2ml of extract, 2ml of dil.ammonium solution was added. b. To the 2ml of extract 2ml thiocyanate solution and 2ml of con HNO ₃ was added	Blood red colour appeared.	Presence of Iron
5.	Test For Zinc: To 2ml of the extract, dil.sodium hydroxide solution was added in 5 drops to excess and dil.ammonium chloride was added.	No White precipitate was formed	Absence of Zinc

6.	Test For Calcium: 2ml of the extract was added with 2ml of 4% dil.ammonium oxalate solution	Cloudy appearance and white precipitate was obtained	Presence of calcium
7.	Test For Magnesium: To 2ml of extract dil.sodium hydroxide solution was added in drops to excess.	No white precipitate was obtained	Absence of Magnesium
8.	Test For Ammonium: To 2ml of extract 1 ml of Nessler's reagent and excess of dil.sodium hydroxide solution are added.	No Brown colour appeared	Absence of ammonium
9.	Test For Potassium: A pinch(25mg) of substance was treated of with 2ml of dil.sodium nitrite solution and then treated with 2ml of dil.cobalt nitrate in 30% dil.glacial acetic acid.	No Yellowish precipitate was obtained.	Absence of Potassium
10.	Test For Sodium: 2 pinches (50mg) of the substance was made into paste by using HCl and introduced into the blue flame of Bunsen burner.	No yellow colour flame appeared	Absence of sodium
11.	Test For Mercury: 2ml of the extract was treated with 2ml of dil.sodium hydroxide solution.	No yellow precipitate was obtained	Absence of mercury
12.	Test For Arsenic: 2ml of the extract was treated with 2ml of dil.sodium hydroxide solution.	No brownish red precipitate was obtained	Absence of arsenic
III. Miscellaneous			
1.	Test For Starch: 2ml of extract was treated with weak dil.iodine solution	Blue colour developed	Presence of starch
2.	Test For Reducing Sugar: 5ml of Benedict's qualitative solution was taken in a test tube and allowed to	Brick red colour developed	Presence of reducing sugar

	boil for 2 minutes and added 8 to 10 drops of the extract and again boil it for 2 minutes. The colour changes are noted.		
3.	Test For The Alkaloids: a) 2ml of the extract was treated with 2ml of dil.potassium iodide solution. b) 2ml of the extract was treated with 2ml of dil.picric acid. c) 2ml of the extract was treated with 2ml of dil.phosphotungstic acid.	Yellow colour developed	Presence of Alkaloid
4.	Test For Tannic Acid: 2ml of extract was treated with 2ml of dil.ferric chloride solution	black precipitate was obtained	Presence of Tannic acid
5.	Test For Unsaturated Compound: To the 2ml of extract 2ml of dil.Potassium permanganate solution was added.	Potassium permanganate was not decolourised	Absence of unsaturated compound
6.	Test For Amino Acid: 2 drops of the extract was placed on a filter paper and dried well. 20ml of Biurette reagent was added.	violet colour developed	Presence of amino acids
7.	Test For Type Of Compound: 2ml of the extract was treated with 2 ml of dil.ferric chloride solution.	No green colour developed No red colour developed No violet colour developed No blue colour developed	Absence of oxy quinole pinephrine and pyro catechol Anti pyrine, Aliphatic amino acids and meconic acid are absent Apomorphine salicylate and Resorcinol are absent Morphine, Phenol cresol and hydro uinone are absent

HIGH PERFORMANCE THIN LAYER CHROMATOGRAPHY

The HPTLC of Takkolathi Chooranam was done at Sri Ramachandra University, Chennai.

Significance of HPTLC fingerprinting in Standardisation

Standardisation of traditional medicine has become mandatory in the present national and international scientific scenario, as they have to stand competing with stringent regulatory methods and also clinically. HPTLC is one of the versatile chromatographic methods presently available for the rapid analysis of herbal drugs due to several reasons. Firstly the time required for the demonstration of the most of the characteristic constituents of a drug is very quick and short. Secondly, in addition to qualitative detection, HPTLC also provides semi-quantitative information on the major active constituents of a drug, thus enabling an assessment of drug quality. Thirdly the fingerprint obtained is suitable for monitoring the identity and purity of drugs and for detecting adulteration and substitution. Hence in order to check the identity, purity and standardise the quantity of active principles in the herbal extracts a HPTLC fingerprint in the formulation has been obtained.

The distribution of phyto-constituents in a plant depends on various factors such as soil, time of collection period of storage, etc. So, it is necessary to standardize the extract being used for pharmacological studies. HPTLC serves as a convenient tool for finding out the distribution pattern of phyto constituents which is unique to each plant. The HPTLC fingerprinting profile establishes the identity and purity of the raw drug being used. It helps in the authentication of the plant material.

Sample Preparation:

100 mg of extract was weighed and dissolved in 70% methanol to get a concentration of 10mg/ml concentration this is then used for injection.

Chromatographic Conditions

The finger printing has been done using the following chromatographic conditions. Chromatography was performed on a 10x10 cm pre activated HPTLC silica gel 60F 254 plate. Samples were applied to the plate as 6mm wide band with an automatic TLC applicator Linomat 5 with N₂ flow (CAMAG, Switzerland), 8mm from the bottom. Densitometric scanning was performed on CAMAG scanner III. The plates were pre-washed by methanol

and activated at 60⁰ C for 5 minutes prior to chromatography. The slit dimension was kept at 5 minutes x 0.45 minutes and 20 minutes scanning speed was employed. The mobile phase was chosen after running each plant in different mobile phases of varying polarity (Toluene, Toluene: Ethyl acetate and Ethyl acetate: Methanol) and 10 ml of mobile phase was used per chromatography. Linear ascending development was carried out in 20 cm x 10-em twin glass chamber saturated with the mobile phase.

Chromatographic Analysis

The hydro alcoholic extracts of the plants have been prepared at a concentration of 10 mg/10 ml in alcohol and were spotted using CAMAG Linomat 5 applicator. The method was optimized by selecting appropriate mobile phase for respective plant extracts and developed in a twin trough chamber, 20 x 10 cm at 25°C. The plates were dried by hair dryer. The developed plates were scanned at appropriate wavelength using CAMAG TLC scanner 3 and photo-documented using CAMAG REPROSTAR 3.

Inferences:

The finger-printing profile establishes the identity and purity of the raw drug being used. It helps in the authentication of the respective plant material. The fingerprinting pattern is characteristic of each plant material used for pharmacological studies. The pattern clearly displays the variation from plant to plant.

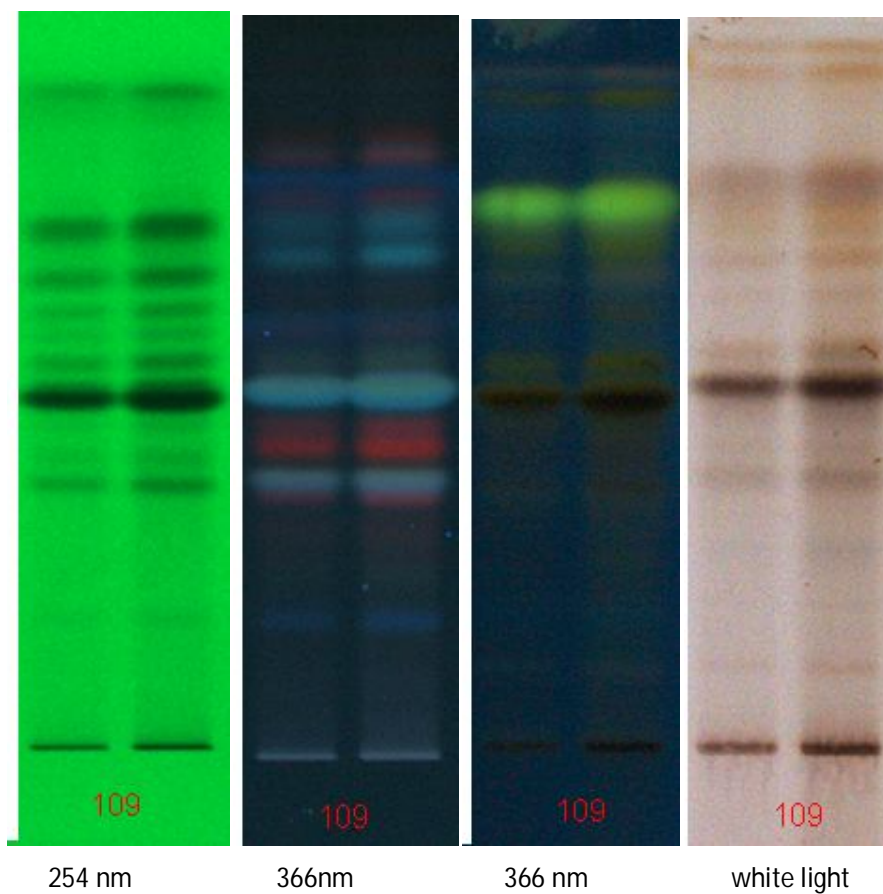
Inference:

HPTLC fingerprint of RH -1 shows four peaks at R_f values 0.25, 0.31, 0.41 & 0.95. The peak correspond to the R_f value 0.31 has maximum peak area of 7256.5. At this stage it is difficult to confirm the individual components present in the extract, but from our lab experience on phytochemical analysis, we suggest that the major peaks found in the fingerprint may be acidic glycosides / resins. Since, in the present chromatographic conditions, the above mentioned components will be eluted easy.

Chromatographic Conditions for HPTLC finger print(Graph 109)

SampleName : Takkolathi chooranam
Sample-ID : 109
Stationary phase : Silica gel F 254
Mobile phase : n-Hexane: Ethyl acetate: Formic acid 60:40:2.5 ml)
Scanning wavelength : 254,298,489 nm
Sample concentration : 20 mg/ml
Injecting volume : 5, 10 µl
Development mode : Ascending mode

HPTLC Fingerprint - RH1



ACUTE AND SUB ACUTE TOXICITY STUDY ON TAKKOLATHI CHOORANAM IN RODENTS

Animals:

Mice of either sex weighing 25-30g and rats weighing 210-240g were obtained from the animal house of Vels University. The animals were used with the approval of the Institute animal ethics committee and obtained from Vels University, Chennai. They were fed with a balanced standard pellet diet and maintained under standard laboratory conditions, providing 24-28⁰C temperature, standard light cycle (12 h light, 12 h dark) and water ad libitum.

Animals were kept in cages with raised floors of wide mesh to prevent coprophagy. Animal welfare guidelines were observed during the maintenance period and experimentation. The rats were randomly assigned to control and different treatment groups, six animals per group. The animals were acclimatized for one week under laboratory conditions.

ACUTE TOXICITY STUDY-OECD 425 GUIDELINES

Acute oral toxicity test for the Takkolathi Chooranam was carried out as per OECD Guidelines 425. As with other sequential test designs, care was taken to ensure that animals are available in the appropriate size and age range for the entire study. The test substance was administered in a single dose by gavage using a stomach tube or a suitable intubation cannula. The fasted body weight of each animal was determined and the dose was calculated according to the body weight. After the substance has been administered, food was withheld for a further 2 hours in mice. The animals were observed continuously for the first 4 h and then each hour for the next 24 h and at 6 hourly intervals for the following 48 h after administering of the test drug, to observe any death or changes in general behaviour and other physiological activities.

Single animals were dosed in sequence usually at 48 h intervals. However, the time interval between dosing is determined by the onset, duration, and severity of toxic signs. Treatment of an animal at the next dose was delayed until one is confident of survival of the previously dosed animal.

Observation of toxicity signs:

General behavior, respiratory pattern, cardiovascular signs, motor activities, reflexes, change in skin and fur, mortality and the body weight changes were monitored daily. The time of onset, intensity, and duration of these signs, if any, was recorded. (Table 1)

SUB-ACUTE TOXICITY

In a 28-days sub acute toxicity study, twenty four either sex rats were divided into four groups of 6 rats each. Group I that served as normal control was administered with distilled water (p.o.) while groups II, III and IV were administered daily with the Takkolathi Chooranam (p.o.) for 28 days at a dose of 100, 200 and 400 g/kg respectively. The animals were then observed daily for gross behavioural changes and any other signs of subacute toxicity.

The weight of each rat was recorded on day 0 and weekly throughout the course of the study, food and water consumption per rat was calculated. At the end of the 28 days they were fasted overnight, each animal was anaesthetized with diethylether, following which they were then dissected and blood samples were obtained by cardiac puncture into heparinised tubes. The blood sample collected from each rat was centrifuged with 3000 X g at 4°C for 10 min to separate the serum and used for the biochemical assays. (Table 2,3,4)

Hematological and blood biochemical analysis:

At the end of the study, all animals were kept fasted for 16-18 h and then anesthetized with anesthetic ether on the 28th day. Blood samples for hematological and blood chemical analyses were taken from retro orbital vein. Heparinized blood samples were taken for determining complete blood count (white blood cell count, differential white blood cell count, platelet count, red blood cell count, hematocrit, and hemoglobin) by semiautomated hematology analyzer.

The serum from non-heparinized blood was carefully collected for blood chemistry and enzyme analysis like glucose, Creatinine, Total protein, Albumin, Total and Direct bilirubins, Serum glutamate-oxaloacetate transaminase (SGOT), Serum glutamate pyruvate transaminase (SGPT), and Alkaline phosphatase (ALP) were automatically determined using autoanalyzer.(Table 5,6,7)

Necropsy:

All rats were sacrificed after the blood collection. The positions, shapes, sizes and colors of internal organs were evaluated. The Spleen, Testes, Pancreas, Lung, Liver, Brain, Heart, Stomach, Intestine, Bone, Ovary, and Kidney tissues were excised from all rats to visually detect gross lesions, and weighed to determine relative organs' weights and preserved in 10% neutral formalin for histopathological assessment. The tissues were embedded in paraffin, and then sectioned, stained with haematoxylin and eosin and were examined microscopically.(Table 8).

Statistical analysis

Values were represented as mean \pm SEM. Data were analysed using one-way analysis of variance (ANOVA) and group means were compared using the Tukey-Kramer Multiple Comparison test using Instat-V3 software. P values < 0.05 were considered significant.

RESULTS:

Clinical signs: Animals were not shown any significant toxic clinical signs during the dosing period of 28 days.

Mortality: All animals from control and all the treated dose groups survived throughout the dosing period of 28 days.

Body weight: Results of body weight determination of animals of control and different dose groups exhibited comparable body weight gain throughout the dosing period of 28 days.

Food consumption: During dosing period, the quantity of food consumed by animals from different dose groups was found to be comparable and normal with that of control animals.

Ophthalmoscopy: Ophthalmoscopic examination of animals in control and test product-treated groups did not reveal any major and remarkable abnormality.

Functional Observations: These tests conducted on the experimental animals at termination and recorded did not reveal any abnormalities.

Urine analysis: Urine analysis data of control group and treated group of animals determined in week 4 did not reveal any abnormalities.

Organ Weight: Comparison of organ weights of treated animals with respective control animals on day 28 was found to be comparable.

Necropsy: Gross pathological examination of animals in control as well as the treated groups did not reveal any abnormalities.

Haematological investigations: The results of haematological investigations conducted on day 28, revealed following significant changes in the values of different parameters investigated when compared with those of respective controls; However, the increase or decrease in the values obtained was within normal biological and laboratory limits.

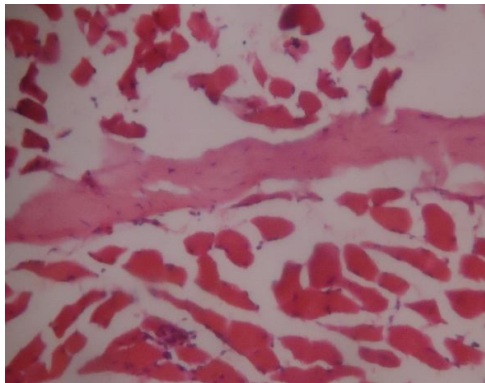
Biochemical Investigations: Results of Biochemical investigations conducted on days 29 revealed significant changes in the values of uric acid and potassium studied when compared with those of respective controls; however, the values obtained were within normal biological and laboratory limits.

CONCLUSION:

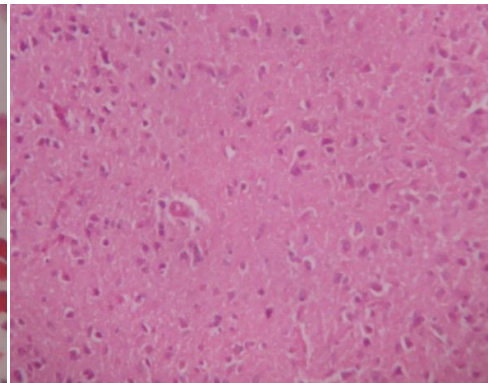
Based on these findings, no toxic effect was observed upto 400mg/kg of Takkolathi Chooranam treated via oral route over a period of 28 days. So, it can be concluded that the Takkolathi Chooranam can be prescribed for therapeutic use in human with the dosage recommendations of upto 400mg/kg. body weight p.o.

**HISTOPATHOLOGICAL SLIDES OF VARIOUS ORGANS AFTER THE
SUBACUTE TOXICITY STUDIES**

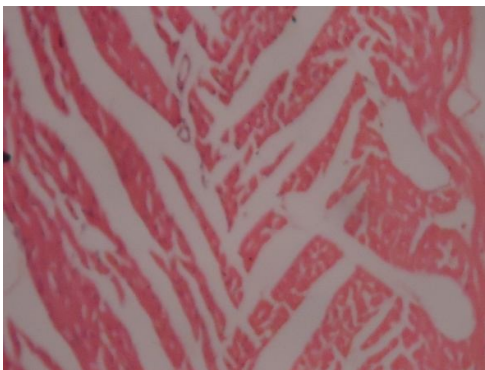
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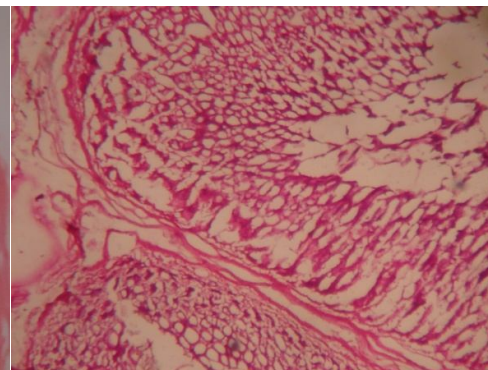
BRAIN



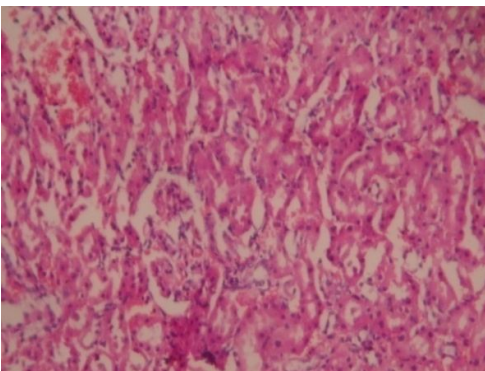
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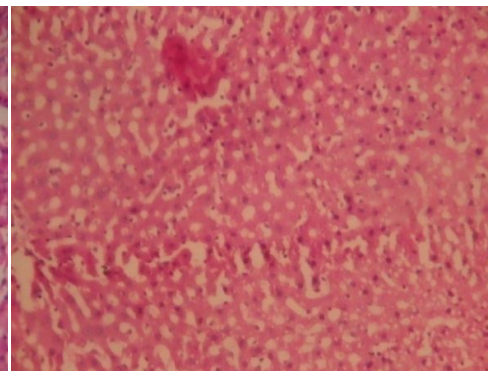
INTESTINE



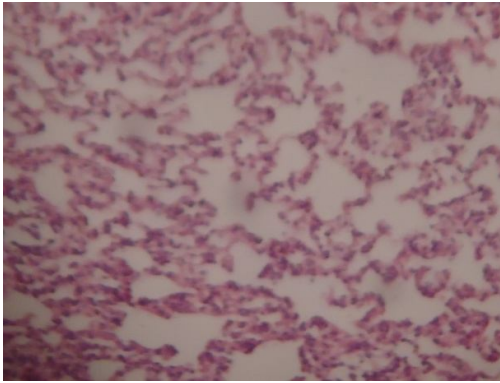
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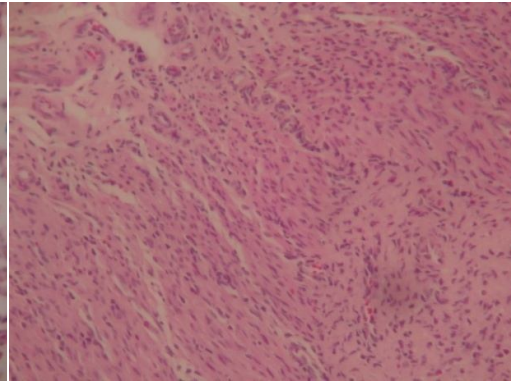
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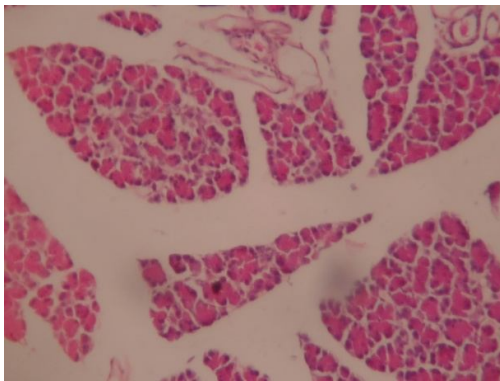
LUNG



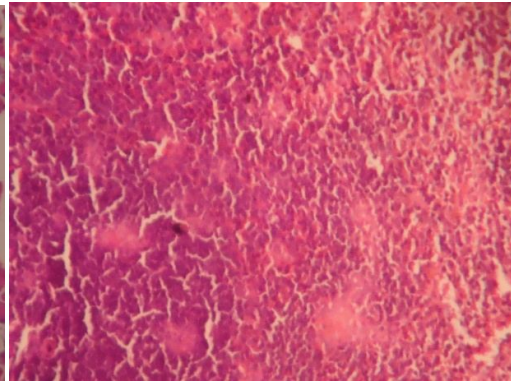
OVARY



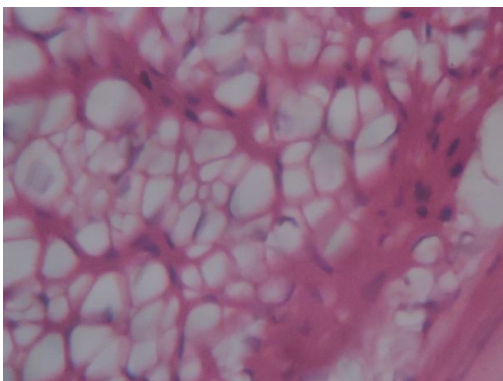
PANCREAS



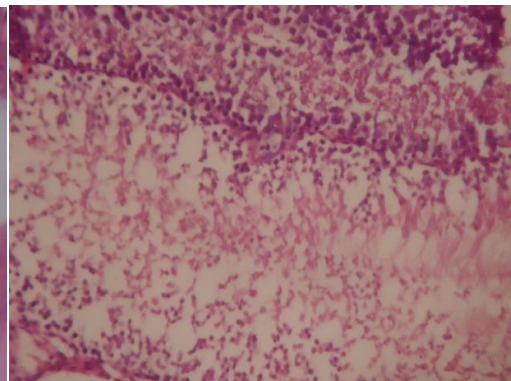
SPLEEN



STOMACH



TESTIS



PHARMACOLOGICAL STUDY OF TAKKOLATHI CHOORANAM

MATERIALS AND METHODS

Histamine solution was freshly prepared in normal saline (NaCl, 8.5 g/l), Carboxy methyl cellulose (2%) (Loba chemie Pvt. Ltd). All other chemicals used were of analytical grade.

Stock solution

The powdered Takkolathi chooranam was mixed uniformly in Carboxy Methyl Cellulose in saline solution to achieve 1mg/ml as main stock solution and used in this study.

Animals

The Guinea pig is a species of rodent belonging to the family Caviidae and the genus Cavia. Despite their common name, these animals are not pigs, nor do they come from Guinea. Guinea pigs of either sex 350- 450 were procured from animal house, Department of Pharmacology, Vels University and throughout the study. Six animals were taken in each group and maintained under controlled environment (temperature $25 \pm 2^{\circ}\text{C}$ and 12 h dark and light cycle) with standard diet and water ad libitum during experiment. All the animals used in this study were approved by CPCSEA. (XIII/VELS/PCOL/34/2000/CPCSEA/IAEC/08.08.2012)

In-vitro Antihistaminic activity

Guinea pig was sacrificed and a segment from ileum (2 cm) was dissected from the terminal ileum and mounted in an organ bath containing Tyrode solution (10 ml) between two stainless steel hooks under 0.5 to 1g initial tension. The lower hook was fixed at the bottom of the organ bath and upper one was connected to an isotonic transducer. The Tyrode solution composition (pH 7.4) was (concentration in gm/lit.) NaCl 8.0, KCl 0.2, CaCl_2 0.2, MgCl_2 0.1, NaHCO_3 1.0, NaH_2PO_4 0.05, and Glucose 1.0gm/liter.

It was continuously aerated and maintained at $37 \pm 0.5^{\circ}\text{C}$ The equilibrium period was 60 min and the bath solution was refreshed every 15 min. After equilibrium period, a dose response curve for histamine in variant molar concentrations, by maintaining 45 min time cycle was used. (Table 1)

Evaluation Of Bronchodilator Activity

Experimental bronchial asthma was induced in guinea pigs by exposing them to histamine. Overnight fasted guinea pigs of either sex (350-450) were selected and randomly divided into four groups each consisting of six animals. Group 1-3 was treated as Test group and received of Takkolathi chooranam at the dose levels of (100, 200, 400 mg/kg). Group 4 was considered as standard and administered with Promethazine (300mg/kg, p.o). All the doses were given orally. Prior to drug treatment each guinea pig was exposed to an atomized fine mist of 2% w/v histamine dihydrochloride aerosol (dissolved in normal saline) using a nebulizer in the histamine chamber. Guinea pigs exposed to histamine aerosol showed progressive signs of difficulty in breathing leading to convulsions, asphyxia and death. The time until signs of convulsion appeared is called pre-convulsion time (PCT) and was determined from the time of exposure to onset of convulsions.

As soon as pre convulsion time was noted, animals were removed from the chamber and placed in fresh air to recover. The percentage protection offered by treatment was calculated by using the following formula: $\text{Percentage protection} = (1 - T_1/T_2) \times 100$. Where; T_1 = the mean of PCT of control group animals. T_2 = the mean of PCT of test group animals.

Statistical analysis

Data were expressed as Mean \pm SEM. Differences between groups were analyzed by one way analysis of variance (ANOVA) followed by Dunnet "t" test. Differences were considered significant when $P < 0.05$ and $P < 0.01$.

RESULTS AND DISCUSSION

Mortality in the acute oral toxicity test was seen in the dose 2000mg/kg. Hence, one-tenth, one twentieth and one fifth of the upper bound dose was considered for the further pharmacological study. Histamine induced bronchoconstriction is the traditional immunological model of antigen induced airway obstruction. Histamine when inhaled causes hypoxia and leads to convulsion in the guinea pigs and causes very strong smooth muscle contraction, profound hypotension and capillary dilation in the cardiovascular system. A prominent effect caused by histamine is severe bronchoconstriction in the guinea pigs that causes asphyxia and death. Histamine was released after degranulation of mast cell by an antigen exposure by antigenic stimulation causing smooth muscle contraction, increased vascular permeability and mucus formation. Histamine is one of the important mediator of

allergy, inflammation and bronchoconstriction. Targeting histamine, either prevention of its release from mast cell or use of histaminergic receptor antagonist becomes part of Antihistaminic therapy in allergic diseases.

In-vivo study of Takkolathi chooranam have been also shown the significant increase in pre-convulsion time due to pre-treatment with Takkolathi chooranam at the dose of 100, 200 and 400mg/kg of body weight of guinea pigs, when the guinea pigs were exposed to histamine. The results of Takkolathi chooranam suggested that it is effective in reducing the symptoms of bronchial asthma and also improve the lung function parameters of asthmatic subjects. The percentage protection of Takkolathi chooranam- 100, 200 and 400 mg/kg, is 16.50, 21.87 and 30.38% respectively.(Table 2)

In the present study, guinea pigs were used because of the extreme sensitivity of their airways to the primary mediators of bronchoconstriction, including histamine and leukotrienes and their ability to be sensitized to foreign proteins. Although there are various model of asthma, guinea pig airways react to histamine, acetylcholine, leukotrienes and other bronchoconstrictors in a manner similar to that seen in humans. Another similarity between the guinea pig model and asthmatic patients is that enhanced bronchoconstriction occurs in both species following sensitization, in response to β -adrenergic antagonists. Thus, the guinea pig model resembles the human allergic pathology in several aspects, especially in terms of mediator release. Histamine antagonists can be conveniently recognized and assayed by their ability to protect guinea pigs against lethal effects of histamine-induced bronchospasm.

CONCLUSION

The Antihistaminic potency on histamine induced contraction of isolated guinea pig ileum was studied and it was observed that the histamine response was decreased in the presence of the test drug Takkolathi chooranam. Histamine when inhaled has been shown to induce bronchoconstriction by direct H_1 -receptor activation and also by a naturally mediated bronchoconstrictor effect via vagal reflexes. The results of present study suggested that Takkolathi chooranam significantly protected the Guinea pigs against histamine-induced bronchospasm. Takkolathi chooranam use traditionally in the management of asthma is justified. The guinea pigs exposed to histamine aerosol showed signs of progressive dyspnoea leading to convulsions. The two doses of Takkolathi chooranam significantly prolonged the latent period of convulsions following the exposure of histamine aerosol. The action started after 1 h of drug administration. Thus, our findings suggest that Takkolathi chooranam possess significant Antihistaminic activity.

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BRONCHIAL ASTHMA¹²

Asthma is an inflammatory disease of the small air-ways, characterized by episodic, reversible bronchial obstruction due to hyper-responsiveness of tracheo-bronchial tree to a multiplicity of intrinsic and extrinsic stimuli manifested clinically by paroxysms of polyphonic wheeze, dyspnoea, and cough which may be relieved spontaneously or as a result of therapy.

Types

- i. Extrinsic Asthma (Atopic Asthma, Early Onset Asthma)
- ii. Intrinsic Asthma (Non-atopic Asthma, late Onset Asthma)

Other types

- i. Nocturnal Asthma
- ii. Gastric Asthma
- iii. Exercise-induces Asthma
- iv. Episodic Asthma
- iii. Chronic Asthma

Factors Precipitating Asthma

- Cold air
- Tobacco smoke
- Dust, acrid fumes
- Emotional stress
- Respiratory infections (viral, bacterial)
- Exercise
- Drugs

NSAIDs especially aspirin

β-blockers

- Chemicals

Sulfiting agents like Na or K bisulfate, sulphur dioxide, etc.

- Allergens

Ingested (fish, nuts, strawberries)

Inhaled (dust, pollen, house dust mite)

Food additives (tartazine, metabisulfite preservatives, monosodium glutamate or ajinomoto)

Occupational allergens (grain-dust, wood-dust).

Pathophysiology:

- Chronic airway inflammation as evidenced by cellular infiltration of airway by activated eosinophils, mast cells, macrophages and T-lymphocytes.
- Released mediators causing bronchial smooth muscle contraction.
- Denudation and desquamation of the epithelium forming mucous plugs that obstruct the airway.

Clinical Features:

- Poly-phonic wheeze
- Dyspnoea
- Cough
- Chest tightness

Differential Diagnosis:

- Chronic Bronchitis
- Emphysema
- Cystic fibrosis
- Pulmonary embolism
- Cardiac failure

Investigations:

- a. Chest X-ray
- b. Pulmonary Function Tests (PFT)
- c. Peak Expiratory Flow Rate (PEFR)

CLINICAL STUDY

Preclinical and clinical study on “TAKKOLATHI CHOORANAM” for “BRONCHODILATOR ACTIVITY” in the management of Eraippu (Bronchial Asthma).
Approval No: NIS/IEC/2011/3/12a

POPULATION AND SAMPLE SIZE:

20 patients of both sexes were enrolled from the Outpatient and Inpatient department of Ayothidass Pandithar Hospital of National Institute of Siddha, Chennai-47.

SUBJECT SELECTION:

INCLUSION CRITERIA:

Age : 18-65 yrs

Sex : both male and female

Weight : 35-85 kgs

Patient having symptoms of

- Wheezing
- Dyspnoea
- Cough with mild expectoration
- Chest tightness

Patient who were willing to provide blood for lab investigation.

Patients who were willing to attend OPD once in 7 days.

Patient who were willing to be admitted in the hospital for 30 days.

Patient willing to sign the informed consent stating that he/she conscientiously stick to the treatment during 30 days but can opt out of the trial of his/her own conscious discretion.

EXCULSION CRITERIA:

- Bronchiectasis
- Pleural effusion
- Pneumonia
- Tuberculosis
- Other type of asthma
- Diabetes mellitus
- Any other serious illness

WITHDRAWAL CEITERIA:

- Development of any adverse reaction
- Occurence of any other serious illness

- Non co-operation of the patient

TREATMENT:

The drug Takkolathi chooranam was administered internally in a dose of 6gm b.d with luke warm water (after food) for 30 days.

CONDUCT OF THE CLINICAL TRIAL:

Bronchial asthma patients satisfying the inclusion criteria were admitted to the trial. Patient was informed about the trial and consent form was obtained . For In-patients, the drug Takkolathi chooranam was administered daily. For Out-patients, the trial drug was issued for seven days course. Weekly clinical assessment was done during review visit to the hospital.

The following investigations were done before and at the end of the treatment.

Blood sample (Hb, TC, DC, ESR, Sugar (F, PP), AEC, T.cholesterol, RFT, LFT)

Peak Expiratory Flow Rate

Urine test (Albumin, Sugar, Deposits)

The following investigations were done before treatment.

X-ray chest - PA view

ECG

Sputum AFB

CLINICAL OBSERVATION:

- ❖ For the clinical study of “Takkolathi Chooranam” on Eraippu, 20 patients were selected.
- ❖ Among 20 patients, 14 (70 %) were in female, 6 (30%) were in male.
- ❖ According to age wise distribution 60% were in 20-35 years, 30% were in 36-50 years and 10% were in 51-65 years.
- ❖ Among 20 patients, All patients were affected from wheeze, 16 patient were affected from dyspnoea, 19 patient were affected with cough, 10 were affected with chest tightness, 17 were affected with expectoration.
- ❖ From the clinical study, 75% patient relieved from wheeze, 75% patient relieved from dyspnoea, 74% patient relieved from cough, 80% patient relieved from chest tightness, 65% patient relieved from expectoration and no adverse effects were observed during trial period.
- ❖ 18 (90%) patients had a significant reduction in the PEFr after the treatment.

DISCUSSION

- The drug Takkolathi Chooranam was selected to evaluate the Bronchodilator and Antihistaminic activity in the management of Eraippu (Bronchial asthma).
- The literary evidence from the text Agathiyar 2000 strongly supports Bronchodilator and Antihistaminic activity of the drug.

Bio-chemical analysis:

Biochemical analysis of the drug Takkolathi choornam reveals the presence of sulphate, iron, calcium, starch, reducing sugar, tannic acid, amino acid and alkaloids.

Iron:

It is necessary for the formation of heme part of hemoglobin. The main function of haemoglobin is the transport of respiratory gases:

- Oxygen from the lungs to tissue
- Carbondioxide from tissues to lungs.

Calcium:

- Calcium is essential for maintaining the integrity of intracellular material.
- It decreases the passages of serum through capillaries in allergic series. Thus calcium is clinically used to reduce allergic exudates.

HPTLC :

- The finger-printing profile establishes the identity and purity of the raw drug being used.
- The fingerprinting pattern is characteristic of each plant material used for pharmacological studies. The pattern clearly displays the variation from plant to plant.

Toxicological studies:

Acute oral toxicity study:

- Mortality in the acute oral toxicity test was seen in the dose 2000mg/kg. Hence, one-tenth, one twentieth and one fifth of the upper bound dose was considered for the further pharmacological study.
- Takkolathi chooranam at the dose of 400mg/kg/po did not exhibit any mortality in rats.

- As per OECD 425 guidelines the dose is said to be “Unclassified” under the toxicity scale. Hence further study with higher doses was not executed.

Sub-acute toxicity for 28 days:

- Based on this study, no toxic effect was observed upto 400mg/kg of Takkolathi Chooranam treated via oral route over a period of 28 days.
- 28 days treatment with the drug did not alter the haematological and biochemical parameters in animals.
- So, it can be concluded that the Takkolathi Chooranam can be prescribed for therapeutic use in human with the dosage recommendations of upto 400mg/kg. body weight p.o.

Pharmacological studies:

- In-vivo study of Takkolathi chooranam have been shown the significant increase in pre-convulsion time (PCT) due to pre-treatment with Takkolathi chooranam at the dose of 100, 200 and 400mg/kg of body weight of guinea pigs.
- The results of study suggested that Takkolathi chooranam significantly protected the Guinea pigs against histamine-induced bronchospasm.
- The study suggested that the trial drug is effective in reducing the symptoms of bronchial asthma and also improve the lung function parameters of asthmatic animal. The percentage protection of Takkolathi chooranam- 100, 200 and 400 mg/kg, is 16.50, 21.87 and 30.38% respectively.
- Hence, pharmacological studies strongly supports that the drug Takkolathi chooranam exhibits significant Antihistaminic activity and Bronchodilator activity

Clinical observation:

- From the clinical study, 75% patient relieved from wheeze, 75% patient relieved from dyspnoea, 74% patient relieved from cough, 80% patient relieved from chest

tightness, 65% patient relieved from expectoration and no adverse effects were observed during trial period.

- 18 (90%) patients had a significant reduction in the PEFr after the treatment.

Bio-statistics:

- Statistically, the paired 't' test shows statistical significance for the symptoms before and after the treatment ($p < 0.0001$).
- Also, the paired 't' test shows statistical significance for the PEFr before and after the treatment ($p < 0.0001$).

Siddha aspect:

- Siddha system of medicine strongly relies on cordial relationship between suvai (taste) and 3 vital forces.
- According to Siddha basic principle, kapha kutram is deranged in Eraippu noi, so the principle aim in the treatment aspect is to make the deranged vital forces into normal by giving the trial drug with karppu suvai and veppa verium.
- Karppu suvai cures inflammatory disease of the throat and removes sputum (phlegm) from the lungs, veppa verium and karppu pirivu pacifies kapham, prapavam of karppu is to correct deranged kapham⁷.

Hence Takkolathi chooranam is a better drug of choice in the management of Eraippu (Bronchial asthma)

SUMMARY

- ❖ The drug Takkolathi chooranam has been selected for this study to evaluate its efficacy on the Bronchodilator and Antihistaminic activity in the management of Eraippu (Bronchial asthma).
- ❖ The literary evidence strongly supports the Bronchodilator and Antihistaminic activity of Takkolathi choornam.
- ❖ The qualitative and quantitative analyses were done at Biochemistry lab, NIS and Sri Ramachandra University, Chennai respectively. Biochemical analysis of the drug Takkolathi choornam reveals the presence of sulphate, iron, calcium, starch, reducing sugar, tannic acid, amino acid and alkaloids.
- ❖ HPTLC was done at Sri Ramachandra University, Chennai. The HPTLC fingerprinting profile establishes the identity and purity of the raw drug used.
- ❖ Preclinical evaluation (acute and sub-acute toxicity study) of the drug was carried out as per OECD guideline in Vels College of pharmacy, Chennai. In the toxicological studies, the drug does not exhibit any mortality upto the dose of 400 mg/kg/po.
- ❖ Preclinical pharmacological study was carried out in animal model in Vels College of Pharmacy, Chennai. In the pharmacological studies, the drug Takkolathi chooranam exhibits significant Anti-histaminic and Bronchodilator activity.
- ❖ As per the Siddha literature and modern science reviews and research articles, the trial drug has potent anti-histaminic and bronchodilator effect.
- ❖ From the clinical study, 75% patient relieved from wheeze, 75% patient relieved from dyspnoea, 74% patient relieved from cough, 80% patient relieved from chest tightness, 65% patient relieved from expectoration and no adverse effects were observed during trial period.

- ❖ 18 (90%) patients had a significant reduction in the PEFV after the treatment.
- ❖ From the statistical analysis-paired 't' test, the drug Takkolathi Chooranam is statistically significant. Statistically, the paired 't' test shows statistical significance for symptoms before and after the treatment. ($p < 0.0001$)
- ❖ The drug Takkolathi Chooranam has
 - Bronchodilator Activity
 - Anti histaminic Activity
 - No side effects
 - No undoing effects
 - Encouraging clinical results.
- ❖ From the clinical and statistical analysis, it is proved that the drug Takkolathi Chooranam has significant Antihistaminic and Bronchodilator activity in the management of Eraippu (Bronchial asthma).

CONCLUSION

- The literary evidence from the text Agathiyar 2000 shows Takkolathi chooranam Antihistaminic and Bronchodilator activity.
- The safety studies (acute toxicity and sub-acute toxicity studies) conducted revealed that the trial drug Takkolathi chooranam is safe. There were no abnormalities found in blood investigation and histo-pathological examination. Hence it can be reasonably assumed that the drug is safe for human use.
- The pharmacological study conducted in animal model shows significant Bronchodilator activity and Antihistaminic activity.
- Clinical study revealed the therapeutic efficacy of the trial drug by showing reduction in Eosinophil, AEC and raised ESR level significantly. There was improvement in other clinical symptoms before and after treatment.
- There was marked improvement in PEFR range after 30 days treatment with Takkolathi chooranam.
- There were no adverse reactions complained during the clinical trial.
- Hence, the drug TAKKOLATHI CHOORANAM can be used in the management of ERAIPPU (Bronchial Asthma).

INTRODUCTION:

Anaemia refers to a state in which the level of hemoglobin and RBCs in the bloodstream is below the normal range approximate for age and sex¹.

Anaemia is a serious public health problem having a direct bearing on physical development, immuno-competence, learning behaviour, maternal mortality, work capacity, GI disturbances, neurocognitive impairment and impaired thermoregulation.

In India and other developing countries iron deficiency anaemia far out numbers all the other types of anaemia out together, as it constitutes 90-95% of the total². It predominantly affects adolescent girls, women of reproductive age and young children⁴.

Iron deficiency anaemia is common among lower socioeconomic group because of insufficient dietary intake and limited absorption of dietary iron.

Iron deficiency anaemia has been universally recognized as the commonest forms of malnutrition occurring in the world. These affect approximately 2 billion people, 80% of whom live in the developing world. One third of world's population has iron deficiency. Southeast Asia has the highest levels at 79%. The Indian subcontinent alone contains nearly half the world's anaemic women³.

According to the National Family Health Survey, anaemia among women between 15-49 years of age is 51.8 percent with 45.7 percent in urban areas and 53.9 percent in rural areas. In studies conducted in developing countries, adolescent anaemia is the greatest nutritional problem. In India, 55 % adolescent girls are anaemic⁴.

The data of operations research, a UNICEF project in Tamil Nadu, documented 95% prevalence of anaemia in Dindigul and 48% in Tirupur among the urban poor³.

Anaemia is correlated with Pandu in Siddha system of medicine. Kumari Parpam is one of the herbo-mineral formulations indicated for Pandu which is mentioned in Siddha text "Agasthiyar 2000", 5th edition, October 2002 by Dr.S.Venkatarajan LIM and published by Saraswathi mahal noolagam⁷. This Parpam has been prepared from Ayam, kandham and gandhagam with the help of Aloe vera juice.

According to Siddha basic principles, function of thuvarpvu suvai is purification of blood and it is also important for RBCs formation. Ayam and gandhagam have thuvarpvu suvai; hence kumari Parpam has immense potential in treating Pandu.

As per Siddha text, Parpam has great efficacy with minimal dosage, sustained availability, stability over long period etc. This Parpam has not been evaluated so far for Pandu, hence the author has selected "Kumari Parpam" to evaluate Haematinic activity.

AIM:

To evaluate the safety and efficacy of Kumari Parpam for Haematinic activity. in the management of Pandu (Anaemia).

OBJECTIVE:**Primary objective:**

To evaluate the Haemetinic activity of Kumari Parpam for Pandu (Anaemia) in preclinical studies..

Secondary objective:

The efficacy of Kumari Parpam has been evaluated in the following aspect.

- Collection of evidences in Siddha and mineralogical aspect
- Biochemical Analysis
- Atomic Absorption Spectrophotometer
- Clinical study- a pilot study on trial medicine

STANDARD OPERATIVE PROCEDURE:

Collection and authentication of the raw drugs:

The raw drug were procured from raw drug store in Chennai and authenticated by competent authority of Department of Gunapadam, National Institute of Siddha, Chennai.

Ingredients:

- | | | |
|-----------------------|---------------------------|-----------------------|
| 1. Purified Ayam | (Iron) | -5 kazhanju(25.5 gm) |
| 2. Purified Kaandham | (magnetic oxide of iron) | -5 kazhanju(25.5gm) |
| 3. Purified Gandhagam | (sulphur) | -5 kazhanju(25.5gm) |
| 4. Katrazhai charu | (Aloe vera juice) | - sufficient quantity |
| 5. Purified Indhuppu | (Sodium chloride impura)* | |
| 6. Purified Chukku | (Zingiber officinale)* | |
| 7. Purified Milagu | (Piper nigrum) * | |
| 8. Purified Thippili | (Piper longum)* | |
| 9. Purified Omam | (Carum copticum)* | |
| 10. Purified Seeragam | (Cuminum cyminum) * | |

* The drugs in s.no 5-10 were taken in equal quantity

Purification process:

Purification of Ayam:

35gm of powder of ayam was soaked in lemon juice for 3 days and then it was washed. Then it was soaked in gingelly oil for 1 day and it was fried in iron vessel. Finally it was soaked in horse gram (*Macrotyloma uniflorum*) decoction for several times⁵.

Purification of Kaandham:

35gm of kaandham was grinded with lemon juice for ¾ hours⁵.

Purification of Gandhagam:

Maruthonri karkam (*Lawsonia inermis*) was mixed with cow's curd and was taken in a mud vessel, a cloth was tied around the mouth of the vessel. The gandhagam was placed over the cloth and it closed with suitable mud vessel. It was then sealed with clay pasted cloth and buried in a pit. Five cow dung cakes were kept over the upper mud vessel and made into pudam, melted gandhagam was collected in the lower mud vessel. Repeat the process for 7 times⁶.

Purification of Indhuppu:

The raw drug was purified by soaking in goat's urine for 3 nazhigai (72 minutes) and dried in the sunlight⁶.

Purification of Chukku:

The raw drug was purified by removing the outerlayer and soaking in the limestone water⁵.

Purification of Milagu:

The raw drug was soaked in buttermilk for 1hour 15 minutes and then it was fried⁵.

Purification of Thippili:

The raw drug was purified by soaking in the lemon juice⁵.

Purification of Omam:

The raw drug was purified by soaking in limestone water for 3 hours and it was fried⁵.

Purification of Seeragam:

The raw drug was kept in sunlight for 6 hours and then it was fried⁵.

Preparation of the medicine:

Purified ayam, kaandham, gandhagam were powdered and grinded with katrazhai charu and made into a villai. The villai was then placed in a mud vessel and it was heated for 4 samam (12 hours) by wooden fuels⁷.

Preparation of the adjuvant:

The chukku, milagu, thippili, indhuppu, omam, seeragam were purified and pulverized by an electric grinder into fine powder, separately. And then it was sieved by using a fine silk cloth (vasthrakayam). The fine powder was purified by pittaviyal method. Then it was dried and ultra filtered by a cotton cloth and made into fine powder again. The powder was stored in a clean, dry air tight glass bottle⁷.

LABELLING:

Name of the preparation	: Kumari Parpam
Colour	: White colour
Dose	: 488 mg b.d
Vehicle	: Honey
Duration	: 1 month
Indication	: Pandu
Date of manufacture	: 13-03-2012
Expiry	: 100 years from the date of manufacture

AYAM:

BEFORE PURIFICATION



AFTER PURIFICATION



GANDHAGAM:

BEFORE PURIFICATION



AFTER PURIFICATION



KAANDHAM:

BEFORE PURIFICATION



AFTER PURIFICATION



VILLAI



ERIPPU



PARPAM



«Ãõ⁶

§ÅÚ | ÅÄ÷, ù:

« ÷, « ÅÍ, « ÅØ, þÊ, þÕõð, ®º | °Ãõ, ÷ÕÍ | ÷, ÷, ÷ÕÀ, ÷Õõð, ÷Õõð, ÷ÕÁ½ð, ÷Õõ | Äý, ÷ÅÍ, ÷ÄØ | ÷, Çõ, ÷¼, °ðÐ, °§Ä | °Ãõ, °Õ¼õ, ¼ÕõÀ, Ðñ ¼õ, Àñ ¼õ, §Ä ÷õ, ÄúâÄ ÷¼õ

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«Ãõ §°Õõ Äìñ Í §¿ìòì ÷É ÁÕõÐ, ù:

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3. « ÄÄì¼Ä Å¼õ¹³

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5.ŞÄİİ Î°òà Äö¹¹

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İö¾ö⁶

ŞÄÜ İÄ÷, ü:

°ÄŞÄİİ §°Äý, ¾Ä½¿ İ ¿İ¾ö, Ý¾ « í İ °ö, ¿ÄŞÄİö ðÄðÆ,
 İÄ°ö¾¿ İ ö Äð¾Ä Äİý, ÓÖý ðÄİ½ö

İÄðİ½ö:

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 ஓரின்றை யாயுளறும் உன்."

İö¾İ ð Äİ ö, İ ýÄö, İÄİ Æ ŞÄö, Äİñ Î , Óö¾İ¼ö, ÄŞİ¾Äö

¿İ½ö

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IRON ^{21, 22, 23}

Chemical Formula: Fe

Empirical formula: Fe⁰⁺

Vernacular names:

Tamil	:	Irimbu
Sans	:	Lauha, Hyam
Malay	:	Basi
Telu	:	Inumu
Urdu	:	Lohchun

Source:

- Widely distributed in both the organic and inorganic kingdoms.
- Found in nearly all rocks, soils, etc, variously combined with oxygen as haematite, with sulphur as iron pyrites.
- Found in the ashes of plants and even the blood (RBC of the blood) of animals and also in the bile, chyle, gastric juice, lymph, milk, pigment of the eye, and in the urine.
- The largest iron resources in the world are in China, Russia, Brazil, Canada, Australia, and India.

Physical properties

- Iron is a silvery-white or grayish metal.
 - It is ductile and malleable.
 - Iron has a very high tensile strength.
- | | | |
|---------------|---|--|
| Meltingpoint | : | 1,536°C(2,797°F) |
| Boilingpoint | : | 3,000°C(5,400°F). |
| Color: | : | Iron black, Dark gray, Steel gray |
| Density: | : | 7.3 - 7.9, Average = 7.6 |
| Diaphaneity: | : | Opaque |
| Fracture: | : | Hackly - Jagged, torn surfaces, (e.g. fractured metals). |
| Hardness: | : | 4-5 - Fluorite-Apatite |
| Luminescence: | : | Non-fluorescent. |
| Luster: | : | Metallic |
| Magnetism: | : | Naturally strong |
| Streak: | : | Gray |

Chemical properties

- Iron is a very active metal. It readily combines with oxygen in moist air. The product of this reaction, iron oxide (Fe_2O_3), is known as rust.
- Iron also reacts with very hot water and steam to produce hydrogen gas. It also dissolves in most acids and reacts with many other elements.

Action:

- Iron improves the quality of blood
- Iron stimulates the functional activity of all the organs of the body and is therefore a valuable general tonic.

MAGNETITE^{24,25}

Chemical Formula: $\text{Fe}^{++}\text{Fe}^{+++}_2\text{O}_4$

Empirical Formula: $\text{Fe}^{3+}_2\text{Fe}^{2+}\text{O}_4$

Physical properties

Cleavage	:	None
Color	:	Grayish black, Iron black.
Density	:	5.1 - 5.2, Average = 5.15
Diaphaneity	:	Opaque
Fracture	:	Sub Conchoidal - Fractures developed in brittle materials characterized by semi-curving surfaces.
Hardness	:	5.5-6 - Knife Blade-Orthoclase
Luminescence	:	Non-fluorescent.
Luster	:	Metallic
Magnetism	:	Naturally strong
Streak	:	black
Curie temperature	:	858 K (585 °C; 1,085 °F).

Chemical properties:

Magnetite reacts with oxygen to produce hematite, and the mineral pair forms a buffer that can control oxygen fugacity.

Source:

Magnetite also occurs in many sedimentary rocks, including banded iron formations.

SULPHUR^{23,26,27}

Chemical Formula : S

Empirical Formula : S₈

Vernacular names:

Tamil	:	Gandakam
Sans	:	Gandhaka
Mal	:	Gendagum
Tel	:	Gandhakam
Hindi	:	Gandak
Eng	:	Brimstone

Source:

- A non-metallic element found free in beds of gypsum and in a state of sublimation in regions of extinct volcanoes ; also in combination with several ores called pyrites, as sulphates and sulphides of iron, copper, lead, zinc, mercury etc.
- In India it occurs naturally in some parts in Nepal, Kashmir, Afghanistan, and in Burma.

Physical properties

Melting point : 115.21 °

Boiling point : 444.6 °C

Color	:	Yellow, Yellowish brown, Reddish, Greenish.
Density	:	2.05 - 2.09, Average = 2.06
Diaphaneity	:	Transparent to translucent
Fracture	:	Sectile - Curved scrapings produced by a knife blade
Hardness	:	1.5-2.5 - Hardness very near Gypsum
Luminescence	:	Non-fluorescent.
Luster	:	Resinous
Streak	:	White

Chemical properties:

- Sulfur burns with a blue flame concomitant with formation of sulfur dioxide, notable for its peculiar suffocating odor.
- Sulfur is insoluble in water but soluble in carbon disulfide and, to a lesser extent, in other nonpolar organic solvents, such as benzene and toluene.

BOTANICAL NAME²⁸: Aloe vera (Linn.) Burm.f;

CLASSIFICATION:

Kingdom	:	Plantae
Subkingdom	:	Tracheobionta
Superdivision	:	Spermatophyta
Division	:	Magnoliophyta
Class	:	Liliopsida
Subclass	:	liliidae
Order	:	liliales
Family	:	Aloaceae
Genus	:	Aloe
Species	:	Aloe vera

VERNACULAR NAMES:

- **Eng.-** Indian aloe, Curacao aloe, Barbados aloe
- **Hindi-** Ghikunwar, Ghikumari;
- **Kan.-** Kathaligida,
- **Mal.-** Kattuvala,
- **Tam.-** Kattalai,
- **Tel.-** Kalabanda

BOTANICAL DESCRIPTION: Leaves radical, inflorescence 90-135 cm long, simple or few branched, Racemes terminal, flowers yellow or orange, about 2.5 cm long, capsules cylindrical.

DISTRIBUTION: Cultivated throughout India, grow wild on the coasts of Bombay, Gujarat and South India.

PARTS USED: Leaf, leaf-juice, dried juice of leaf

ACTIONS : The plant is bitter, sweet, cooling, anthelmintic, aperients, carminative, deobstruent, depurative, diuretic, stomachic, emmenagogue, ophthalmic.

Physical constants: Foreign matter-Not more than 2%; Total ash-Not more than 5 %; Acid insoluble ash-Not more than 2%; Alcohol soluble extractive- Not less than 60 %; Moisture

content – Not more than 10 % of its weight when dried to constant weight at 105 degree Celsius.

CHEMICAL CONSTITUENTS: Hydroxyanthraquinone-barbaloin and γ -hydroxyaloin isomers. The other constituents include aloe emodin, chrysophanol, chromone derivatives – aloeresin B with its p-coumaryl derivatives oleoresin A and C and the aglycone aloesone.

ACTION: Anti inflammatory, hepatoprotective, antibacterial and cathartic.

ADULTERANTS: Marketed drug may be adulterated with black catechu [Acacia catechu Wild.], pieces of irons and stones.

PHYSICAL PROPERTIES OF KUMARI PARPAM

The physical properties of Kumari Parpam was carried out at Sri Ramachandra University, Chennai.

pH at 10% of aqueous solution:

Five grams of Kumari Parpam was weighed accurately and placed in clear 100 ml beaker. Then 50 ml of distilled water was added to it and dissolved well. After 30 minutes it was then applied in to pH meter at standard buffer solution of 4.0, 7.0, 9.2.

Ash Values

The Ash values are a measure of the inorganic constituents present in the raw drug. A high ash content explains its unsuitable nature to be used as a drug.

Total Ash

A little of extract was taken in a silica crucible previously ignited, cooled and weighed. It was incinerated by gradually increasing the heat not exceeding dull red heat (450°C) until free from carbon, cooled and weighed. The percentage of ash was calculated with reference to air-dried drug. The procedure was repeated to get the constant weight.

Water soluble ash

The total ash was boiled with 25 ml water and filtered through ash less filter paper (Whatmann 4.1). It was followed by washing with hot water. The filter paper was dried and ignited in the silica crucible, cooled and the water insoluble ash was weighed. The water-soluble ash can be calculated by subtracting the water insoluble ash from the total ash.

Acid insoluble ash

The total ash obtained was boiled for 5 minutes with 25 ml of (10% w/v) dilute hydrochloric acid and filtering through ash less filter paper (Whatmann 4.1). The filter paper was ignited in the silica crucible, cooled and insoluble ash was weighed. (Table B)

BIO -CHEMICAL ANALYSIS OF KUMARI PARPAM

The biochemical analysis of the Kumari Parpam was carried out in the Biochemistry lab, NIS.

S.No	EXPERIMENT	OBSERVATION	INFERENCE
1.	Appearance of sample	white in colour	
2.	Solubility: a. A little(500mg) of the sample was shaken well with distilled water. b. A little(500mg) of the sample was shaken well with con. HCl/Con. H ₂ SO ₄	Sparingly soluble	Absence of Silicate
3.	Action of Heat: A small amount(500mg) of the sample was taken in a dry test tube and heated gartly at first and then strong.	No white fumes evolved	Absence of Carbonate
4.	Flame Test: A small amount(500mg) of the sample was made into a paste with con. HCl in a watch glass and introduced into non-luminous part of the Bunsen flame.	No Bluish green flame appeared.	Absence of Copper
5.	Ash Test: A filter paper was soaked into a mixture of sample and dil. cobalt nitrate solution and introduced into the Bunsen flame and ignited.	Yellow colour flame appeared.	Presence of sodium

Preparation of Extract:

5gm of Kumari Parpam was weighed accurately and placed in a 250ml clean beaker and added with 50ml of distilled water. Then it was boiled well for about 10 minutes. Then it was cooled and filtered in a 100ml volumetric flask and made up to 100ml with distilled water.

S.No	EXPERIMENT	OBSERVATION	INFERENCE
	I. Test For Acid Radicals		
1.	Test For Sulphate: a. 2ml of the above prepared extract was taken in a test tube to this added 2ml of 4% dil ammonium oxalate solution b. 2ml of the above prepared extracts was added with 2ml of dil-HCl was added until the effervescence ceases off. Then 2ml of dil. Barium chloride solution was added.	Cloudy appearance present	Presence of Sulphate
2.	Test For Chloride: 2ml of the above prepared extract was added with dil. HCl till the effervescence ceases. Then 2ml of dil. silver nitrate solution was added.	Cloudy appearance present	Presence of Chloride
3.	Test For Phosphate: 2ml of the extract was treated with 2ml of dil. ammonium molybdate solution and 2ml of con. HNO ₃ .	No Yellow appearance.	Absence of Phosphate
4.	Test For Carbonate: 2ml of the extract was treated with 2ml dil. Magnesium sulphate solution	No Cloudy appearance.	Absence of carbonate
5.	Test For Nitrate: 1gm of the substance was heated with copper turning and concentrated H ₂ SO ₄ and viewed the test tube vertically down.	No Brown gas evolved.	Absence of Nitrate
6.	Test For Sulphide: 1gm of the substance was treated with 2ml of con. HCL	No Rotten Egg Smelling gas.	Absence of Sulphide
7.	Test For Fluoride & Oxalate: 2ml of extract was added with 2ml of dil. Acetic acid and 2ml dil. calcium chloride solution and heated.	No cloudy appearance present	Absence of fluoride and oxalate
8.	Test For Nitrite:	No Characteristic	Absence

	3drops of the extract was placed on a filter paper, on that-2 drops of dil.acetic acid and 2 drops of dil.Benzidine solution was placed.	changes	of Nitrite
9.	Test For Borate: 2 Pinches(50mg) of the substance was made into paste by using dil.sulphuric acid and alcohol (95%) and introduced into the blue flame.	No Bluish green colour flame.	Absence of borate
	II. Test For Basic Radicals		
1.	Test For Lead: 2ml of the extract was added with 2ml of dil.potassium iodine solution.	No yellow Precipitate obtained.	Absence of Lead
2.	Test For Copper: a. One pinch(50mg) of substance was made into paste with con. HCl in a watch glass and introduced into the non-luminous part of the flame.	No Blue colour flame No Blue colour precipitate formed.	Absence of copper
3.	Test For Aluminium: To the 2ml of extract, dil.sodium hydroxide was added in 5 drops to excess.	No Yellow colour appeared.	Absence of aluminium
4.	Test For Iron: a. To the 2ml of extract, 2ml of dil.ammonium solution was added. b. To the 2ml of extract 2ml thiocyanate solution and 2ml of con HNO ₃ was added	Blood red colour appeared.	Presence of Iron
5.	Test For Zinc: To 2ml of the extract, dil.sodium hydroxide solution was added in 5 drops to excess and dil.ammonium chloride was added.	No White precipitate was formed	Absence of Zinc
6.	Test For Calcium: 2ml of the extract was added with 2ml of 4% dil.ammonium oxalate solution	Cloudy appearance and white precipitate was obtained	Presence of calcium
7.	Test For Magnesium: To 2ml of extract dil.sodium	No White precipitate was	Absence of Magnesium

	hydroxide solution was added in drops to excess.	obtained	
8.	Test For Ammonium: To 2ml of extract 1 ml of Nessler's reagent and excess of dil.sodium hydroxide solution are added.	No Brown colour appeared	Absence of ammonium
9.	Test For Potassium: A pinch(25mg) of substance was treated of with 2ml of dil.sodium nitrite solution and then treated with 2ml of dil.cobalt nitrate in 30% dil.glacial acetic acid.	No Yellowish precipitate was obtained.	Absence of Potassium
10.	Test For Sodium: 2 pinches (50mg) of the substance was made into paste by using HCl and introduced into the blue flame of Bunsen burner.	yellow colour flame appeared	Presence of sodium
11.	Test For Mercury: 2ml of the extract was treated with 2ml of dil.sodium hydroxide solution.	No yellow precipitate was obtained	Absence of mercury
12.	Test For Arsenic: 2ml of the extract was treated with 2ml of dil.sodium hydroxide solution.	No brownish red precipitate was obtained	Absence of arsenic
III.Miscellaneous			
1.	Test For Starch: 2ml of extract was treated with weak dil.iodine solution	No blue colour developed	Absence of starch
2.	Test For Reducing Sugar: 5ml of Benedict's qualitative solution was taken in a test tube and allowed to boil for 2 minutes and added 8 to 10 drops of the extract and again boil it for 2 minutes. The colour changes are noted.	Brick red colour not developed	Absence of reducing sugar

3.	Test For The Alkaloids: a) 2ml of the extract was treated with 2ml of dil.potassium iodide solution. b) 2ml of the extract was treated with 2ml of dil.picric acid. c) 2ml of the extract was treated with 2ml of dil.phosphotungstic acid.	Yellow colour developed	Presence of Alkaloid
4.	Test For Tannic Acid: 2ml of extract was treated with 2ml of dil.ferric chloride solution	No black precipitate was obtained	Presence of Tannic acid
5.	Test For Unsaturated Compound: To the 2ml of extract 2ml of dil.Potassium permanganate solution was added.	Potassium permanganate was not decolourised	Absence of unsaturated compound
6.	Test For Amino Acid: 2 drops of the extract was placed on a filter paper and dried well. 20ml of Biurette reagent was added.	violet colour developed	Presence of amino acids
7.	Test For Type Of Compound: 2ml of the extract was treated with 2 ml of dil.ferric chloride solution.	No green colour developed No red colour developed No violet colour developed No blue colour developed	Absence of oxy quinole pinephrine and pyro catechol Anti pyrine, Aliphatic amino acids and meconic acid are absent Apomorphine salicylate and Resorcinol are absent Morphine, Phenol cresol and hydro uinone are absent

ATOMIC ABSORPTION SPECTROPHOTOMETER ON KUMARI PARPAM

Atomic Absorption Spectrophotometer of Kumari Parpam was carried out at Sri Ramachandra University, Chennai.

Elemental Analysis using Atomic Absorption Spectrophotometer:

The elemental analysis of digested samples have been determined by Atomic Absorption Spectrophotometer (AAS model 400 Perkin Elmer) at the Sri Ramachandra University, Chennai. The element Fe has been analyzed. In this method the sample, in the form of a homogeneous liquid, is introduced into a flame where thermal and chemical reactions create “free” atoms capable of absorbing, emitting or fluorescing at characteristic wavelengths.

In Atomic Absorption Spectrophotometer (AAS) the majority of free atoms in the commonly used flames were in the ground state, but that the flames did not also have enough energy to excite these atoms. A light source emitting a narrow spectral line of the characteristic energy is used to excite the free atoms formed in the flame. The decrease in energy (absorption) is then measured.

In AAS the wave length (nm), Flame type, Atomizer, Measurement mode, Lamp source and calibration range (ppm) of different elements have been used, are listed in table.

Instrumental conditions for elemental analysis

Parameter	Fe
Wavelength (nm)	248.3
Flame composition	A-Ac
Atomizer	Standard Burner Head
Measurement mode	Absorbance
Calibration Range (ppm)	0.04 - 2
Lamp source	HCL

A - Ac: Air-Acetylene; HCL: Hallow cathode lamp; EDL: Electrode less discharge lamp

METHODOLOGY

I. Microwave Digestion For Elemental Analysis

Model Name: Multiwave3000

Digestion Procedure:

200mg of the given sample is placed in a digestion vessel, acid is added and the mixture is heated for several minutes. After the digestion, the samples are diluted to a specific volume. If too much sample is used in wet digestion, the reaction mixture can become violent. The samples are placed in digestion vessels that fit directly into digestion racks. There are several different acids or mixtures of acids used for digestion, the most common of which is concentrated Hydrochloric acid. The samples are heated slowly at a high temperature. After digestion, the samples are diluted to the appropriate volume with deionized H₂O.

II. Elemental Analysis using Atomic Absorption Spectrophotometer

The elemental analysis of digested samples have been determined by Atomic Absorption Spectrophotometer- Flame technique (AAS model 400 Perkin Elmer). Working standard solutions of Fe was prepared from stock standard solution of 1000 ppm from MERCK. Using blank solution to zero the instrument performs the Calibration. The standards are then analyzed and their absorbance recorded. A graph of Absorbance Vs Concentration is plotted. Calibration of the instrument was repeated periodically during operation. A blank reading was also taken and necessary correction was made during the calculation of concentration of various elements. (Table C)

The digested material was made upto 100 ml for analysis in an (AAS) atomic absorption spectrophotometer (Perkin Elmer). The results were calibrated using standard calibration curve.

In AAS the Wave Length (nm), Flame type, Lamp source and Calibration range (ppm) of different elements have been used, are listed in table.

Instrumental conditions for elemental analysis

Element	Wavelength nm	Light source	Flame type
Iron	386	HCL	Air/Ac

Air/Ac: Air-Acetylene; HCL: Hallow cathode lamp

ACUTE AND SUB ACUTE TOXICITY STUDY ON KUMARI PARPAM IN RODENTS

Animals:

Mice of either sex weighing 25-30g and rats weighing 210-240g were obtained from the animal house of Vels University. The animals were used with the approval of the Institute animal ethics committee and obtained from Vels University, Chennai. They were fed with a balanced standard pellet diet and maintained under standard laboratory conditions, providing 24-28⁰C temperature, standard light cycle (12 h light, 12 h dark) and water ad libitum. Animals were kept in cages with raised floors of wide mesh to prevent coprophagy. Animal welfare guidelines were observed during the maintenance period and experimentation. The rats were randomly assigned to control and different treatment groups, six animals per group. (Approval number: XIII/VELS/PCOL/35/2000/CPCSEA/IAEC/08.08.2012).. The animals were acclimatized for one week under laboratory conditions.

ACUTE TOXICITY STUDY-OECD 425 GUIDELINES

Acute oral toxicity test for the Kumari Parpam was carried out as per OECD Guidelines 425. As with other sequential test designs, care was taken to ensure that animals were available in the appropriate size and age range for the entire study. The test substance was administered in a single dose by gavage using a stomach tube or a suitable intubation cannula. The fasted body weight of each animal was determined and the dose was calculated according to the body weight. After the substance has been administered, food was withheld for a further 2 hours in mice. The animals were observed continuously for the first 4 h and then each hour for the next 24 h and at 6 hourly intervals for the following 48 h after administering of the test drug, to observe any death or changes in general behaviour and other physiological activities. Single animals were dosed in sequence usually at 48 h intervals. However, the time interval between dosing was determined by the onset, duration, and severity of toxic signs. Treatment of an animal at the next dose was delayed until one was confident of survival of the previously dosed animal.

Observation of toxicity signs:

General behavior, respiratory pattern, cardiovascular signs, motor activities, reflexes, change in skin and fur, mortality and the body weight changes were monitored daily. The time of onset, intensity, and duration of these signs, if any, was recorded (Table 1).

SUB-ACUTE TOXICITY:

In a 28-days sub acute toxicity study, twenty four either sex (3+3) rats were divided into four groups of 6 rats each. Group I that served as normal control was administered with distilled water (p.o.) while groups II, III and IV were administered daily with the Kumari Parpam (p.o.) for 28 days at a dose of 50, 100, 200 mg/kg respectively. The animals were then observed daily for gross behavioural changes and any other signs of subacute toxicity. The weight of each rat was recorded on day 0 and weekly throughout the course of the study, food and water consumption per rat was calculated. At the end of the 28 days they were fasted overnight, each animal was anaesthetized with diethylether, following which they were then dissected and blood samples were obtained by cardiac puncture into heparinised tubes. The blood sample collected from each rat was centrifuged with 3000 X g at 4°C for 10 min to separate the serum and used for the biochemical assays. (Table 2,3,4)

Hematological and blood biochemical analyses:

At the end of the study, all animals were kept fasted for 16-18 h and then anesthetized with anesthetic ether on the 28th day. Blood samples for hematological and blood chemical analyses were taken from retro orbital vein. Heparinized blood samples were taken for determining complete blood count (white blood cell count, differential white blood cell count, platelet count, red blood cell count, hematocrit, and hemoglobin) by semiautomated hematology analyzer. The serum from non-heparinized blood was carefully collected for blood chemistry and enzyme analysis (glucose, blood urea nitrogen (BUN), creatinine, total protein, albumin, total and direct bilirubins, serum glutamate-oxaloacetate transaminase (SGOT), serum glutamate pyruvate transaminase (SGPT), and alkaline phosphatase (ALP)) were automatically determined using autoanalyzer. (Table 5,6,7).

Necropsy:

All rats were sacrificed after the blood collection. The positions, shapes, sizes and colors of internal organs were evaluated. The Spleen, Testes, Pancrea, Lung, Liver, Brain, Heart, Stomach, Intestine, Bone, Ovary, and Kidney tissues were excised from all rats to visually detect gross lesions, and weighed to determine relative organs' weights and preserved in 10% neutral formalin for histopathological assessment. The tissues were embedded in paraffin, and then sectioned, stained with haematoxylin and eosin and were examined microscopically.(Table 8)

Statistical analysis:

Values were represented as mean \pm SEM. Data were analysed using one-way analysis of variance (ANOVA) and group means were compared using the Tukey-Kramer Multiple

Comparisons Test using GraphPad InStat-V3 software. P values < 0.05 were considered significant.

RESULTS

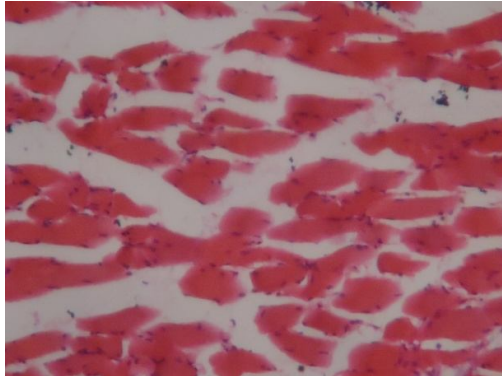
- 1) All the animals from control and all the treated dose groups up to 200 mg/kg survived throughout the dosing period of 28 days.
- 2) No signs of major or significant intoxication were observed in animals from lower to higher dose groups during the dosing period of 28 days.
- 3) Animals from all the treated dose groups exhibited comparable body weight gain with that of controls throughout the dosing period of 28 days.
- 4) Food consumption of control and treated animals was found to be comparable throughout the dosing period of 28 days.
- 5) Ophthalmoscopic examination, conducted prior to and at the end of dosing period on animals from control and all the treated dose groups did not reveal any abnormality.
- 6) Haematological analysis conducted at the end of the dosing period on day 28, revealed significant abnormalities attributable to the treatment with higher dose.
- 7) Biochemical analysis conducted at the end of the dosing period on day 28, revealed remarkable abnormalities in urea level.
- 8) Functional observation tests conducted at termination revealed no major abnormalities.
- 9) Urine analysis, conducted at the end of the dosing period in week 4 and at the end of recovery period, revealed no abnormality.
- 10) Organ weight data of animals sacrificed at the end of the dosing period was found to be comparable with that of respective controls.
- 11) Gross pathological examination did not reveal any significant abnormality.
- 12) Histopathological examination did not reveal any abnormality.

CONCLUSION:

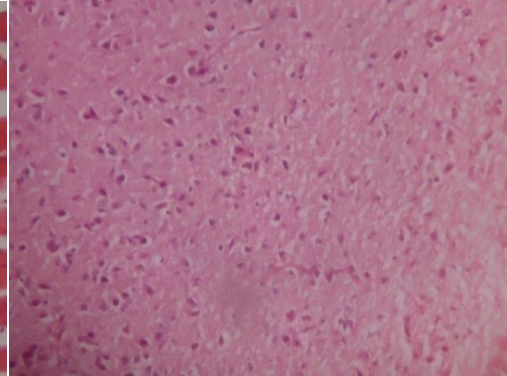
Based on these findings, it was decided as no toxic effect was observed upto 200 mg/kg of Kumari Parpam treated via oral route over a period of 28 days. So, it can be concluded that the Kumari Parpam can be prescribed for therapeutic use in human with the dosage recommendations of upto 200 mg/kg. body weight p.o.

**HISTOPATHOLOGICAL SLIDES OF VARIOUS ORGANS AFTER THE
SUBACUTE TOXICITY STUDIES**

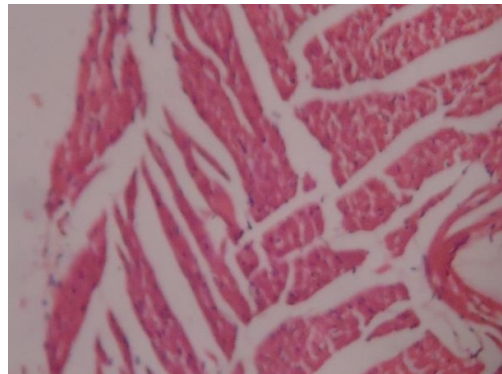
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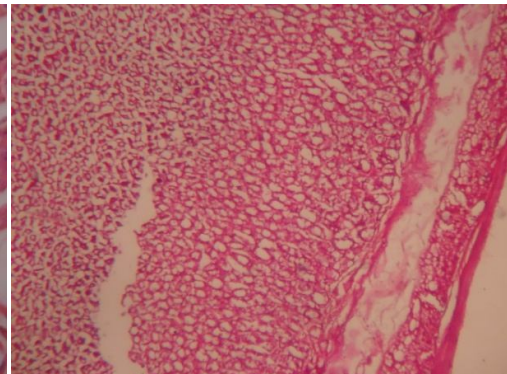
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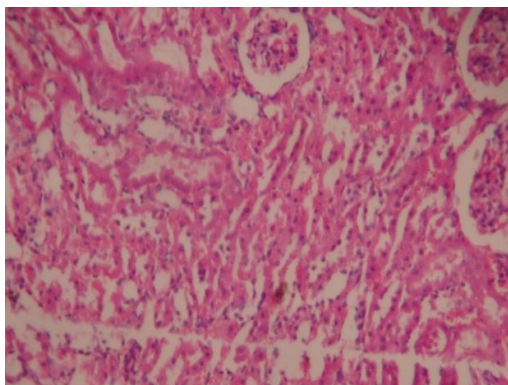
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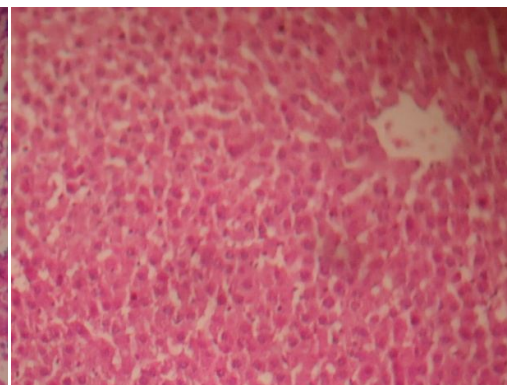
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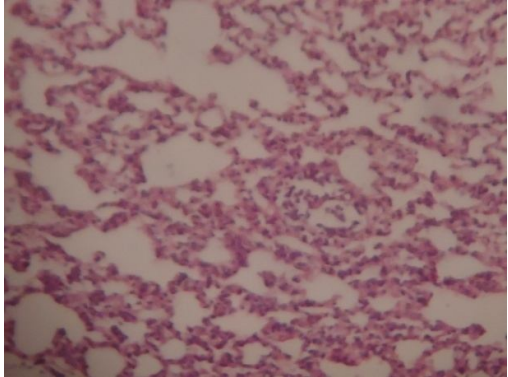
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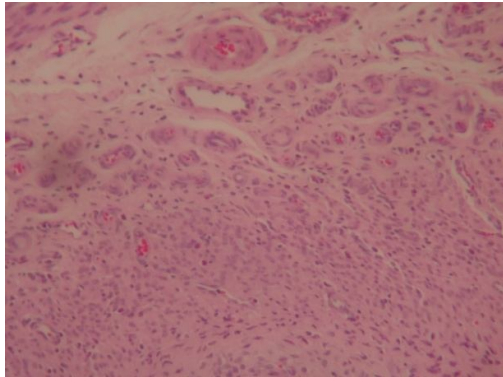
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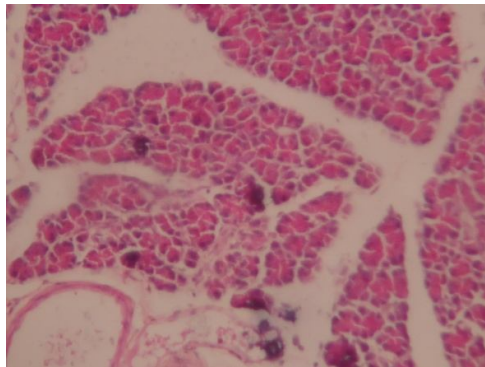
LUNG



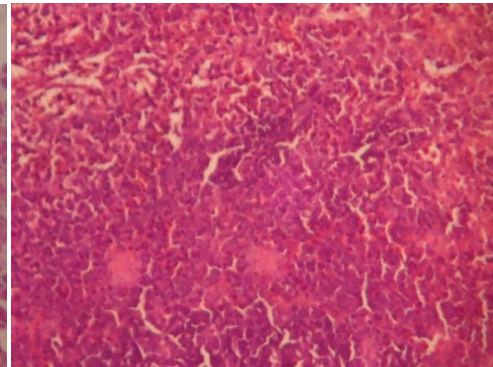
OVARY



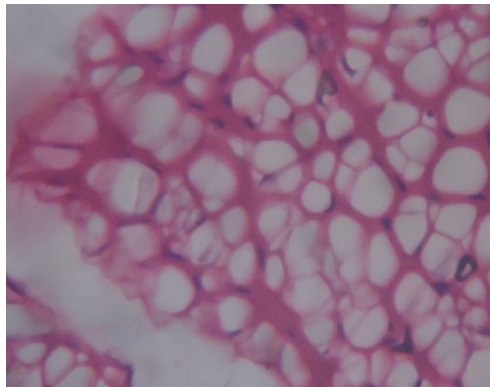
PANCREAS



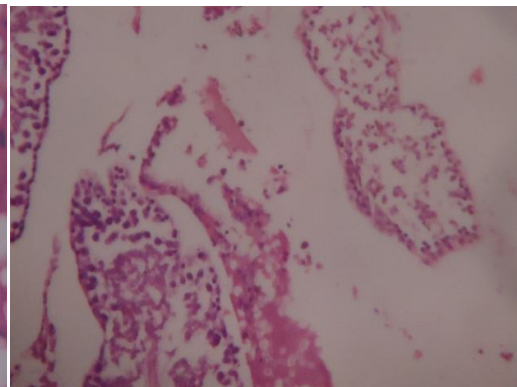
SPLEEN



STOMACH



TESTIS



PHARMACOLOGICAL STUDY ON KUMARI PARPAM

MATERIALS AND METHODS:

Drug and Stock solution

The Kumari Parpam was prepared as per the procedure in traditional Siddha text recommendation and made into suspension form using CMC as a suspending agent and used in this study. The resulting suspension was then grounded and filtered. The filtrate was stored in a refrigerator until use. The suspension was further diluted with 2% CMC so as to achieve 200mg/ml stock concentration.

Animals:

Male albino rats (150-180g) and Mice of either sex weighing 25-30g were used for the study. The rats were housed in wire-mesh cages with a 12 h light/dark cycle. They had continuous access to food and water during the entire period of experimentation. (Approval number: XIII/VELS/PCOL/35/2000/CPCSEA/IAEC/08.08.2012).

Evaluation of Haematinic Activity :

Six rats were kept as normal control group (Group 1), while 24 rats were made anaemic by oral intubations of phenylhydrazine (10 mg/kg body weight) daily for seven days. Rats that developed anaemia with haemoglobin concentration <14 g/dl were recruited for the study. Anaemic rats were randomly divided into 5 groups (2 to 6) and treated as follows: Group 1: received distilled water (1 ml) daily (normal control), Group 2: received 2% CMC (1 ml) daily (anaemic control), Group 3: received oral single dose of the Kumari Parpam 50 mg/kg body weight/day Group 4: received oral single dose of the Kumari Parpam 100 mg/kg, Group 5: received oral single dose of the Kumari Parpam 200 mg/kg Group 6: received oral single dose of the heamatinic syrup 2ml/kg body weight/day. The treatment was continued for 2 weeks.

Haematological investigation:

Before and after treatment with drug Kumari Parpam blood was collected from the retro orbital vein of experimental animals after an overnight fast (T=0) and after 1 and 2 weeks of treatment with Kumari Parpam, was used for the determination of red blood cell count (RBC), haemoglobin (Hb) concentration and packed cell volume (PCV). The mean cell volume (MCV), mean cell haemoglobin (MCH) and the mean cell haemoglobin concentration (MCHC) were calculated.

Statistical analysis

Experimental data was analysed using analysis of variance (ANOVA) and Dunnet's 't' test to determine significant differences between means. The statistical analysis system (INSTAT-V3) package was used for this analysis.

RESULTS AND DISCUSSION

Mortality in the acute oral toxicity test was seen in the dose 1000mg/kg. Hence, one-tenth, one twentieth and one fifth of the upper bound dose was considered for the further pharmacological study. It has been reported that phenyl hydrazine causes oxidative damage to red cells by increasing the formation of reactive oxygen species.

The changes in the haematological parameters of the rats during the study are tabulated. The RBC, Hb, and PCV of rats administered Phenylhydrazine decreased significantly ($P<0.01$) while the MCV and MCH increased giving rise to macrocytic anaemia ($P<0.05$). Kumari Parpam at the dose of 50, 100, 200 mg/kg showed good percentage of improving in haemoglobin level, which was almost equivalent to standard treated group indicating correction of anaemia induced by Phenyl hydrazine after 14 days treatment.

Treatment with Kumari Parpam at the dose levels 100 and 200mg/kg for 14 day showed significant increase in RBC ($p<0.01$) compared to positive control and it was comparable to standard drug used in this study. Phenylhydrazine altered the haematological parameters by haemolysis characterized by decrease in haemoglobin concentration, total RBC counts and PCV on day 7. However, the haematological parameters were restored to normal range after treatment with Kumari Parpam for 14 days. Effective changes was observed after one week of treatment of anaemic rats with Kumari Parpam reversed the influence of Phenylhydrazine resulting to a significant ($P<0.05$) increase in RBC, Hb, and PCV. The Hb, RBC and PCV reached near normal at the second week of the treatment.

Rats treated with Phenylhydrazine (10mg/kg/day for 7 days) resulted in a marked haemolytic anaemia characterised by decreased RBC, Hb and PCV. The main function of the RBC is the transportation of oxygen in to the tissues of the body. At such, any pathological or physiological condition that affects the RBC alters its function and this may be detrimental to the body. In this study Phenylhydrazine altered the function of RBC by haemolysis characterised by decreased levels of RBC, Hb and PCV. However, this effect was restored after one week of Kumari Parpam treatment. Also the recovery was progressive such that after 1week of continuous treatment, the Hb concentration and PCV were higher in the treated groups than in the normal control group.

CONCLUSION

In order to provide effective, safe and cheap drug and to prove the traditional claim for the treatment of anemic conditions the Kumari Parpam at a dose of 50, 100 and 200 mg/kg (p.o.), was evaluated and found significantly increased the haemoglobin, haematocrit, and RBC count in anaemic rats indicating the Haematinic effect. The rapid and progressive recovery of anaemic rats responding to treatment of Kumari Parpam may be due to increased erythropoiesis. However, the mechanism of action by which Kumari Parpam produced its effect on increasing RBC, Hb and PCV in experimental animals need to be evaluated in a scientific manner using specific experimental animal models and also clinical trials are required to understand the exact molecular mechanisms of action. Based on the results it can be concluded that the Kumari Parpam is a good drug of choice for the anemia at the dose level of 100 mg/kg. (Table 1,2, 3).

ANAEMIA¹⁰

Anaemia may be defined as a state in which the blood haemoglobin level is below the normal range for the patient's age and sex (Males < 12g/dL; females < 10g/dL).

Causes of Anaemia:-

- Decreased or ineffective marrow production
 - a. Inadequate iron, B₁₂ or folate, trace elements (zinc, cobalt)
 - b. Hypoplasia of bone marrow
 - c. Infiltration by malignant cells

- Peripheral causes (increases RBC destruction or loss)
 - a. Blood loss
 - b. Haemolysis
 - c. Hypersplenism

Classification of Anaemia:-

- i. Microcytic Anaemias (MCV < 80 fL)
- ii. Macrocytic Anaemias (MCV > 100 fL)
- iii. Normocytic Anaemias (MCV 80-100 fL)

Iron Deficiency Anaemia

Haemoglobin is normally the largest iron compartment of the body. Hb is 0.34% iron by weight. In an adult, total iron content of Hb compartment is about 2 gm.

Iron Metabolism

Iron taken in diet is absorbed at all parts of GI tract especially duodenal mucosa. Acid medium favours iron absorption. Acid medium favours formation of soluble macromolecular complexes of iron with vitamin C, sugar, amino acid or bile in the duodenum. Only 10% of the ingested iron is absorbed. Normal serum iron level is 50 to 150 mg/dl.

Frank iron deficiency increases absorption by 30 – 40% and in iron overload, absorption decrease.

- Iron absorption is increased in the below:
 - i. Ferrous state

- ii. Increases erythropoiesis
- iii. Iron deficiency.
- Iron absorption is decreased in below:
 - i. Ferric state
 - ii. In the presence of phosphates and phytates
 - iii. Bone marrow hypoplasia.

The absorbed iron is stored in the form of ferritin (water soluble form) and haemosiderin (water insoluble form)

Causes of Iron Deficiency

1. Increased iron utilization (increased demand)
 - Postnatal growth spurt
 - Adolescent growth spurt
 - Erythropoietin therapy
2. Physiologic iron loss
 - Menstruation
 - Pregnancy
3. Pathologic iron loss
 - Gastrointestinal bleeding
 - Genitourinary bleeding
 - Pulmonary haemosiderosis
 - Intravascular haemolysis
 - Phlebotomy for polycythaemia rubra vera
4. Decrease iron intake
 - Cereal rich diet
 - Pica, food fads, malabsorption
 - Acute or chronic inflammation.

Stages in Iron Deficiency Anaemia

There are three stages in the development of iron deficiency anaemia.

1. Negative iron balance
2. Iron deficient erythropoiesis
3. Iron deficient anaemia.

Clinical Features

Patients may have

- angular stomatitis
- atrophic glossitis
- koilonychia
- brittle hair
- pruritis
- pica
- Plummer-Vinson syndrome(postcricoid web) or
- menorrhagia.

Investigations:

- Hemoglobin
- Hematocrit value (PCV)
- MCV,MCH, MCHC
- Peripheral smear study
- Serum ferritin
- Total iron binding capacity
- Serum transferrin
- Serum Iron

CLINICAL STUDY

Preclinical and clinical study on “KUMARI PARPAM” for “HAEMATINIC ACTIVITY” in the management of Pandu (Anaemia).

Approval No. NIS/ IEC/ 2011/ 3/ 12b

POPULATION AND SAMPLE SIZE:

20 patients of both sexes were enrolled from the Outpatient and Inpatient department of Ayothidass Pandithar Hospital of National Institute of Siddha, Chennai-47.

SUBJECT SELECTION:

INCLUSION CRITERIA:

Age : 18-65 yrs

Sex : both male and female

Weight : 35-85 kgs

Patient having symptoms of

- Tiredness
- Dyspnoea on exertion
- Giddiness
- Headache
- Insomnia
- Palpitation
- Poor appetite

Patient who are willing to provide blood for lab investigation.

Patients who are willing to attend OPD once in 7 days.

Patient willing to sign the informed consent stating that he/she conscientiously stick to the treatment during 30 days but can opt out of the trial of his/her own conscious discretion.

EXCULSION CRITERIA:

- Pregnancy and lactation
- Known case of cirrhosis and chronic renal failure
- Patient receiving Anti-tuberculosis drugs
- Known case of Hypothyroidism
- Chronic blood loss
- Any other serious illness

WITHDRAWAL CRITERIA:

- Development of any adverse reaction
- Occurrence of any other serious illness
- Non co-operation of the patient

TREATMENT:

The drug Kumari Parpam was administered internally in a dose of 488 mg b.d with honey (after food) for 30 days.

CONDUCT OF THE CLINICAL TRIAL:

Anaemia patients satisfying the inclusion criteria were admitted to the trial. Patient was informed about the trial and consent form was obtained. For Out-patients the trial drug was issued for seven days course. Weekly clinical assessment was done during review visit to the hospital.

The following investigations were done before and at the end of the treatment.

- Blood sample (Hb, TC, DC, ESR, HCT/PCV, MCV, MCH, MCHC, and smear study, Sugar (F, PP), AEC, T.cholesterol, RFT and LFT)
- Urine test (Albumin, Sugar, Deposits)
- Motion test (ova, cyst, albumin)

CLINICAL OBSERVATION:

- For the clinical study of “Kumari Parpam” on Pandu, 20 patients were selected.
- Among 20 patients, 18(70 %) were in female, 2(30%) were in male.
- According to age wise distribution 30% were in 20-35 years, 55% were in 36-50 years and 15% were in 51-65 years.
- Among 20 patients, 18 patients were pallor, 13 patients were affected from breathlessness, 20 patients were affected from tiredness, 19 patients were affected with giddiness, 18 patients were affected from anorexia, 6 patients were affected with pica, and 15 were affected with palpitation.
- From the clinical study, 61% patient relieved from pallor, 69% patient relieved from breathlessness, 60% patient relieved from tiredness, 63% patient relieved from giddiness, 66.7% patient relieved from anorexia, 66.7% patient relieved from pica, 66.7% patient relieved from palpitation and no adverse effects were observed during trial period.

DISCUSSION

- The drug Kumari Parpam was selected to find out the Haematinic activity in the management of Pandu (Anaemia).
- The literary evidence from the text Agathiyar 2000 strongly supports Haematinic activity of the drug.

Bio-chemical analysis:

Biochemical analysis of the drug Kumari Parpam reveals the presence of sulphate, sodium, chloride, iron, calcium, tannic acid, amino acid and alkaloids.

Iron:

- It helps in the formation of heme, which is joined with globin to form the haemoglobin.
- It is one of the RBC maturation factor.

Calcium:

- It increases iron absorption from GIT.

Sulphate:

- It is useful for the absorption of iron from mucosa.

Atomic Absorption Spectrophotometer:

- The elemental analysis of digested sample has been determined by Atomic Absorption Spectrophotometer.
- By AAS, Iron content (Fe ppm) in the trial drug was calculated as 0.132ppm. This strongly supports the Haematinic activity of the drug Kumari Parpam.

Toxicological studies:

Acute oral toxicity study:

- Mortality in the acute oral toxicity test was seen in the dose 1000mg/kg. Hence, one-tenth, one twentieth and one fifth of the upper bound dose was considered for the further pharmacological study.
- As per OECD 425 guidelines the dose is said to be “Unclassified” under the toxicity scale. Hence further study with higher doses was not executed.

Sub-acute toxicity for 28 days:

- Based on this study, no toxic effect was observed upto 200 mg/kg of Kumari Parpam treated via oral route over a period of 28 days.
- 28 days treatment with the drug did not alter the haematological and biochemical parameters in animals.

- So, it can be concluded that the Kumari Parpam can be prescribed for therapeutic use in human with the dosage recommendations of upto 200 mg/kg. body weight p.o.

Pharmacological studies:

- Kumari Parpam at the dose of 50, 100 and 200 mg/kg showed good percentage of improvement in haemoglobin level, which was almost equivalent to standard treated group indicating correction of anaemia induced by Phenyl hydrazine after 14 days treatment.
- The Kumari Parpam at a dose of 50, 100 and 200 mg/kg (p.o.), was evaluated and found significantly increased the haemoglobin, haematocrit, and RBC count in anaemic rats indicating the Haematinic effect.
- Based on the results it can be concluded that the Kumari Parpam is a good drug of choice for the anemia at the dose level of 100mg/kg.

Clinical observation:

- From the clinical study, 61% patient relieved from pallor, 69% patient relieved from breathlessness, 60% patient relieved from tiredness, 63% patient relieved from giddiness, 66.7% patient relieved from anorexia, 66.7% patient relieved from pica, 66.7% patient relieved from palpitation and no adverse effects were observed during trial period.

Bio-statistics:

- Statistically, the paired 't' test shows statistical significance for the symptoms before and after the treatment. ($p < 0.0001$).
- Also, the paired 't' test shows statistical significance for the Hb level before and after the treatment ($p < 0.0001$).

Siddha aspect:

- Siddha system of medicine strongly relies on cordial relationship between suvai and 3 vital forces.
- The principle aim in the treatment aspect is to make the deranged vital forces into normal by giving the trial drug that has thuvorppu suvai.
- According to Siddha basic principles, function of thuvorppu suvai is the purification of blood and it is also important for RBCs formation.

Hence KUMARI PARPAM is a better drug of choice in the management of PANDU (Anaemia).

SUMMARY

- ❖ The drug Kumari Parpam has been selected for this study to evaluate its efficacy on the Haematinic activity in the management of Pandu (Anaemia).
- ❖ The literary evidence strongly supports the Haematinic activity of Kumari Parpam.
- ❖ The qualitative and quantitative analysis were done at Biochemistry lab, NIS and Sri Ramachandra University, Chennai respectively. Biochemical analysis of the drug Kumari Parpam reveals the presence of sulphate sodium, chloride, iron, calcium, tannic acid, amino acid and alkaloids.
- ❖ The elemental analysis of sample has been determined by Atomic Absorption Spectrophotometer and it was done at Sri Ramachandra University, Chennai.
- ❖ Preclinical evaluation (acute and sub-acute toxicity study) of the drug was carried out as per OECD guideline in Vels College of pharmacy, Chennai. In the toxicological studies, the drug does not exhibit any mortality upto the dose of 400 mg/kg/po.
- ❖ Preclinical pharmacological study was carried out in animal model in Vels College of Pharmacy, Chennai. In the pharmacological studies, the drug Kumari Parpam exhibits significant Haematinic activity.
- ❖ From the clinical study, 61% patient relieved from pallor, 69% patient relieved from breathlessness, 60% patient relieved from tiredness, 63% patient relieved from giddiness, 66.7% patient relieved from anorexia, 66.7% patient relieved from pica, 66.7% patient relieved from palpitation and no adverse effects were observed during trial period.
- ❖ From the statistical analysis-paired 't' test, the drug Kumari Parpam is statistically significant. Statistically, the paired 't' test shows statistical significance for symptoms before and after the treatment. ($p < 0.0001$)
- ❖ The drug Kumari Parpam has
 - Haematinic activity
 - No side effects
 - No undoing effects
 - Encouraging clinical results.
- ❖ From the clinical and statistical analysis it is proved that the drug KUMARI PARPAM is statistically significant on Haematinic activity in the management of PANDU (Anaemia).

CONCLUSION

- The literary evidence from the text Agathiyar 2000 shows Kumari Parpam has Haematinic activity.
- The safety studies (acute toxicity and sub-acute toxicity) studies conducted revealed that the trial drug Kumari Parpam is safe. There were no abnormalities found in blood investigation and histo-pathological examination. Hence it can be reasonably assumed that the drug is safe for human use.
- From Atomic Absorption Spectrophotometer, iron content of the trial drug, was calculated as 0.132ppm, which shows its Haematinic activity.
- The pharmacological study conducted in animal model shows significant Haematinic activity.
- Clinical study revealed the therapeutic efficacy of the trial drug by showing raise in Haemoglobin, Haematocrit value, TRBC significantly. There was improvement in other clinical symptoms before and after treatment.
- There were no adverse reactions complained during the clinical trial.
- Hence, the drug KUMARI PARPAM can be used in the management of PANDU (Anaemia).

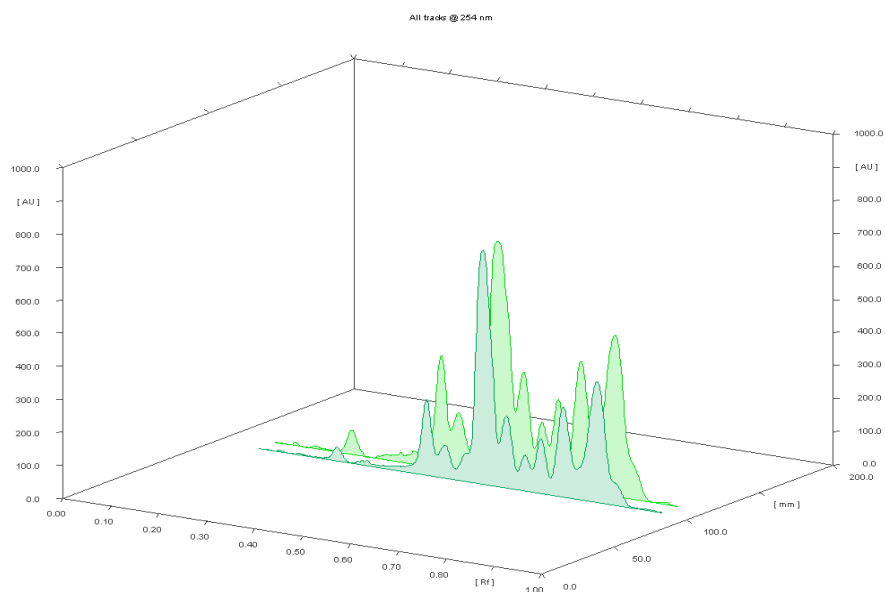
PHYSICAL PROPERTIES: (Table A)

S.NO	Characteristic test	Results
1	pH	5.06
2	Ash Value	0.95
3	Water soluble ash	0.00
4	Acid insoluble ash	0.04

QUALITATIVE ANALYSIS:

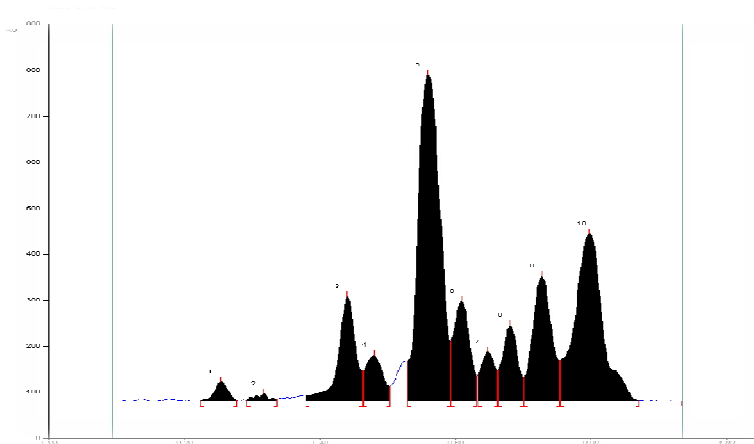
S.NO	PARAMETERS	RESULTS
1	Calcium	Present
2	Sulphate	Present
3	Magnesium	Absent
4	Iron	Present
5	Aminoacids	Absent
6	Starch	Present
7	Flavonoids	Absent
8	Phosphate	Absent
9	Tannic acid	Present
10	Glucose	Present

109 – HPTLC Profile

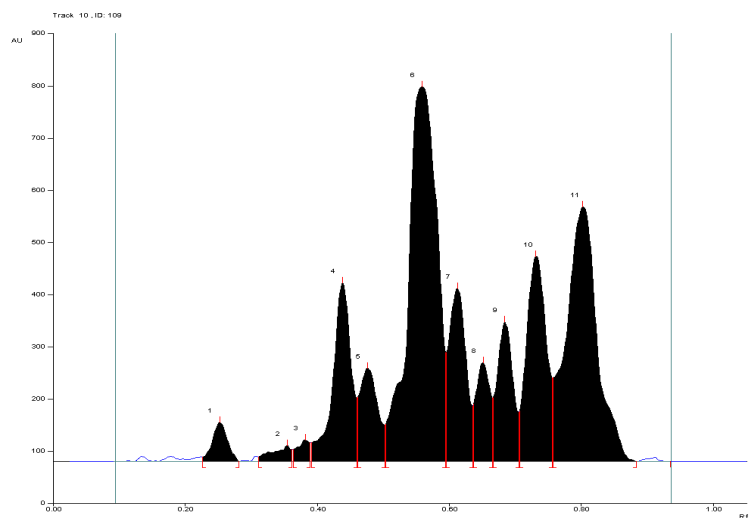


254nm

254nm 3D display

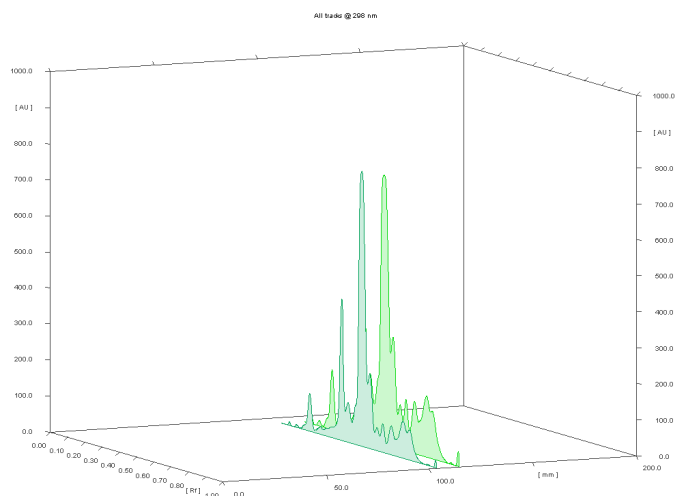


5µl (254nm)

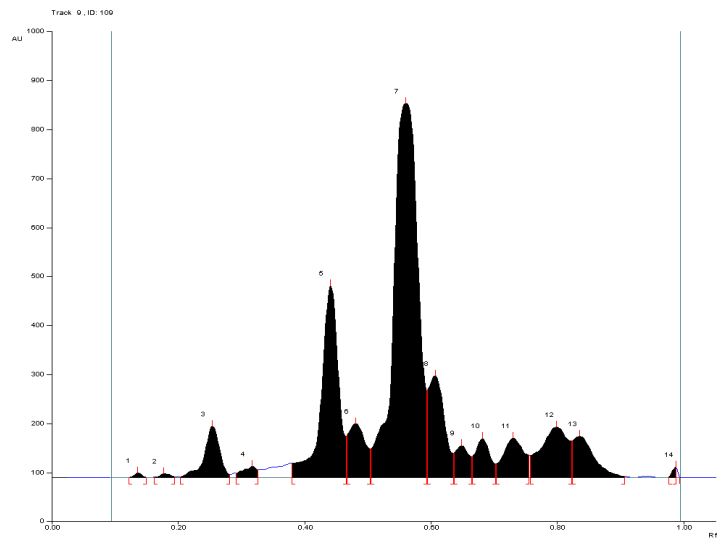


10 µl (254nm)

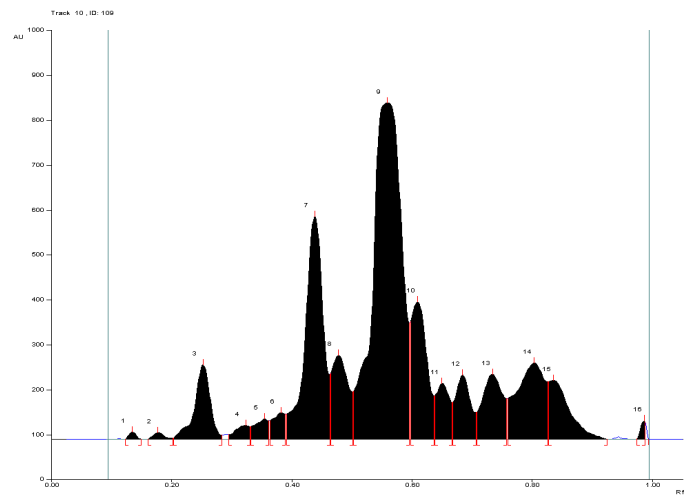
298nm



298 nm 3D display

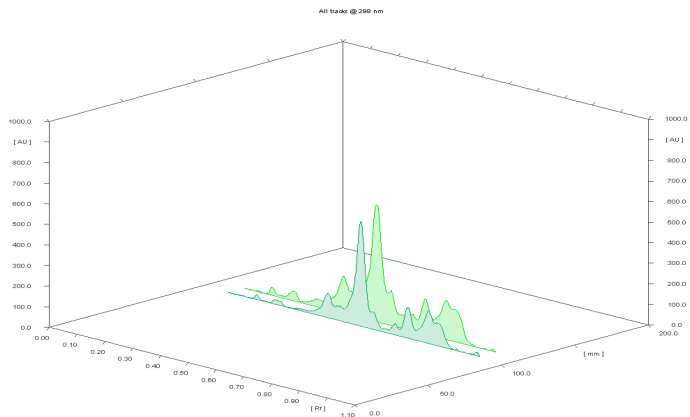


5µl (298nm)

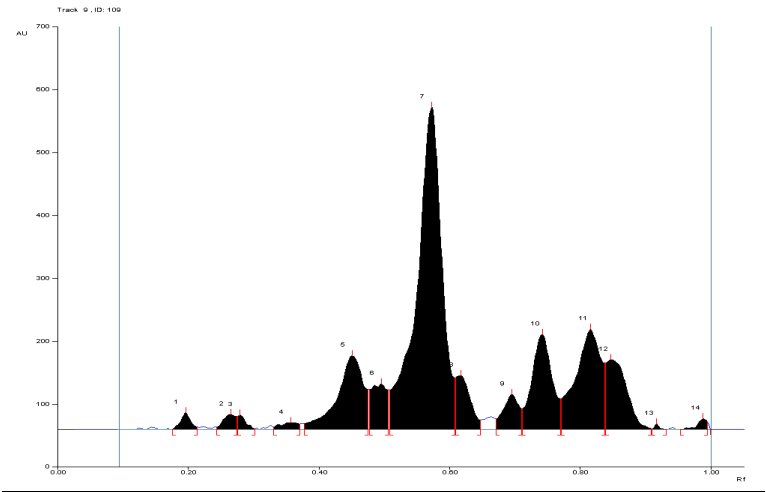


10µl (298nm)

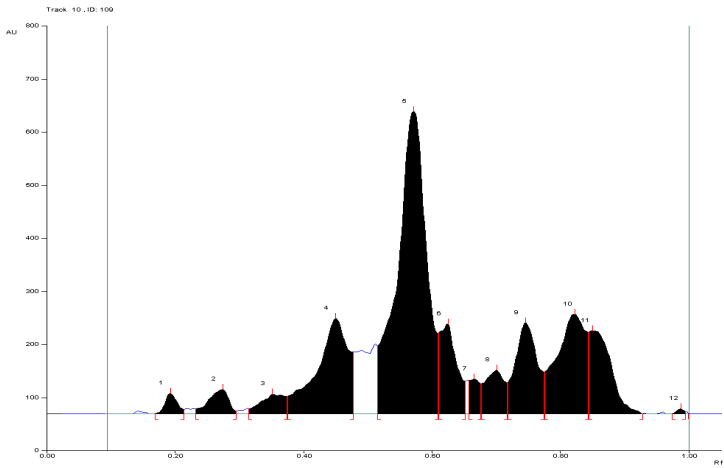
Derivatisation (298nm)



298 nm 3D display

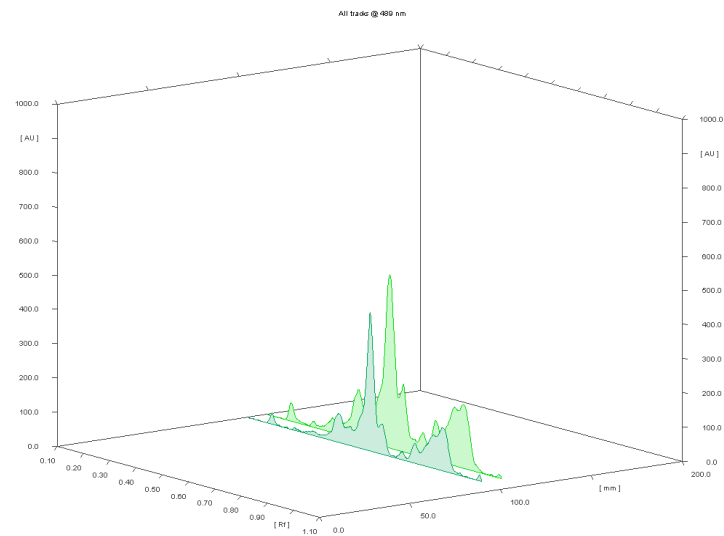


Derivatisation 5µl (298nm)

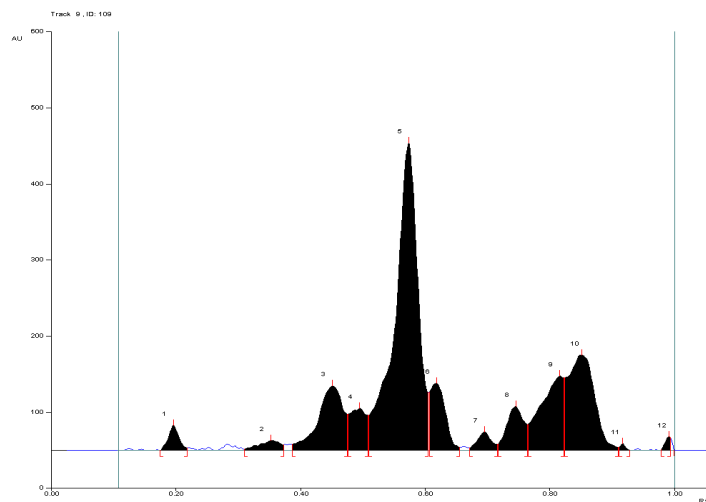


Derivatisation 10µl (298nm)

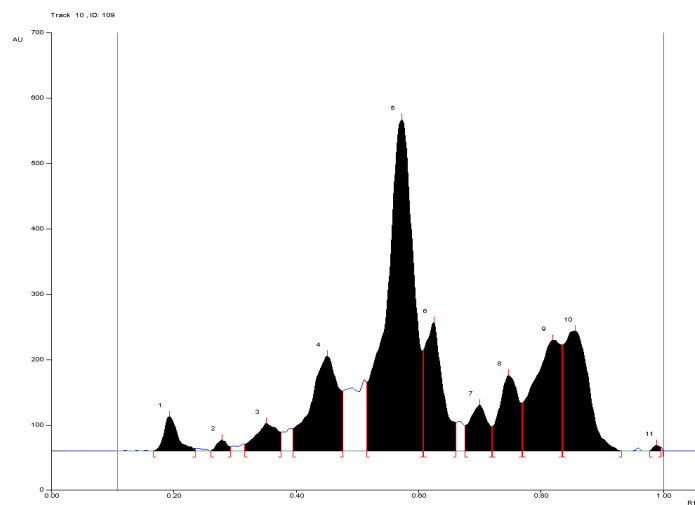
Derivatisation (498nm)



489 nm 3D display



Derivatisation 5µl (498nm)



Derivatisation 10µl (498nm)

Fingerprint chromatogram of RH -1 at 404nm

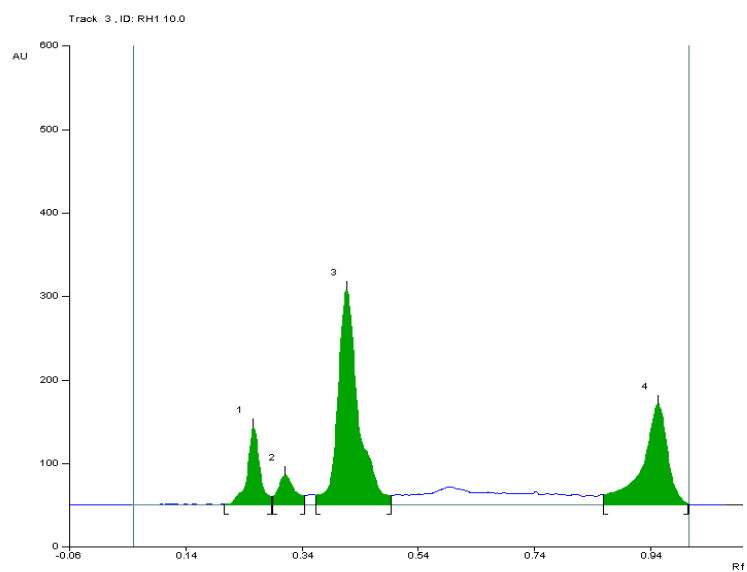


Table 1: Dose finding experiment and its behavioral Signs of Toxicity

No	Dose mg/kg	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
1	500	+	-	-	+	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-
2	1000	+	-	-	+	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-
3	2000	+	-	-	+	-	+	-	-	-	-	-	-	-	-	-	-	+	-	-	-

1. Alertness 2. Aggressiveness 3. Pile erection 4. Grooming 5. Gripping 6. Touch Response 7. Decreased Motor Activity 8. Tremors 9. Convulsions 10. Muscle Spasm 11. Catatonia 12. Muscle relaxant 13. Hypnosis 14. Analgesia 15. Lacrimation 16. Exophthalmos 17. Diarrhoea 18. Writhing 19. Respiration 20. Mortality

Table 2. Body wt (g) of albino rats exposed to Takkolathi Chooranam for 28days.

Dose (mg/kg/day)	Days				
	1	7	14	21	28
Control	124.72±4.22	122.62±5.12	125.10±5.12	127.12±4.88	128.12±5.00
100	126.15±4.16	125.30±4.00	128.20±5.13	131.00±7.15	135.21±6.46
200	123.35±3.46	125.61±5.52	125.87±4.54	128.32±5.10	129.00±7.25
400	132.20±5.77	134.11±5.15	136.42±4.82	135.19±5.43	135.20±8.00

Values are mean ± S.E.M. (Dunnet't' test). ^{ns}P > 0.05; Vs Control N=6.

Table 3. Food (g/day) intake of rats exposed to Takkolathi Chooranam for 28days.

Dose (mg/kg/day)	Days (gms/rats)				
	1	7	14	21	28
Control	46.12±2.40	45.36±2.72	45.98±2.28	45.08±2.28	47.11±3.46
100	44.40±2.56	45.12±2.44	45.58±2.36	47.31±2.45	48.31±3.85
200	42.15±2.48	42.45±2.88	46.22±2.54	46.99±3.00	46.21±3.12
400	44.06±2.77	45.20±2.76	47.30±2.66	48.02±2.54	48.64±3.02

Values are mean ± S.E.M. (Dunnet't' test). ^{ns}P > 0.05; Vs Control N=6.

Table 4. Water (ml/day) intake of rats exposed to Takkolathi Chooranam for 28days.

Dose (mg/kg/day)	Days(ml/rat)				
	1	7	14	21	28
Control	50.12±2.48	51.64±3.58	52.46±3.10	52.11±3.23	53.00±3.35
100	51.56±2.55	51.12±3.10	51.00±3.69	52.46±3.18	54.55±2.46
200	51.24±2.96	53.10±3.05	52.22±3.44	52.36±2.82	55.12±3.21
400	50.48±3.02	52.82±3.12	50.12±3.02	51.55±3.12	55.66±3.12

Values are mean ± S.E.M. (Dunnet't' test). ^{ns}P > 0.05; Vs Control N=6.

Table 5. Hematological parameters after 28days treatment with Takkolathi Chooranam in rats.

Parameter	Control	100 mg/kg	200 mg/kg	400 mg/kg
RBC (millions/cu.mm)	5.21±0.38	5.10±0.30	5.48±0.36	5.98±0.28
Hb (g/dl)	13.25±0.31	13.56±1.1	13.32±1.4	13.10±1.2
PCV (%)	45.02±1.46	44.2±2.0	44.58±2.2	45.12±2.4
WBC(cells/cu.mm)	7375±440.17	7425±329.2	7224±325.1	7458±425.2
Neutrophil (%)	52.45±4.22	54.21 ±3.21	49.34±3.14	48.64±3.42
Lymphocytes (%)	40.00±2.23	41.2±3.1	47.21±3.2	48.11±3.5
Eosinophil's (%)	6.24±0.66	5.68±0.54	5.95±0.45	5.77±0.42
Monocytes (%)	4.1±0.2	3.2±0.2*	3.4±0.3	3.3±0.2
Basophils (%)	0±0	0±0	0±0	0±0
Platelets (10⁵ cells/cu.mm)	1.46±0.06	1.55±0.07	1.78±0.05**	1.46±0.04
MCV(Fl)	79.2±2.0	81.5±1.5	80.1±2.1	82.2±2.4
MCHC (pg)	25.2±1.2	24.00±1.4	25.2±1.0	26.05±1.5

Values are mean of 6 animals ± S.E.M. (Dunnett's test). *P<0.05; **P<0.01. N=6.

Table 6. Effect of treatment with Takkolathi Chooranam biochemical (LFT, RFT, and Lipid Profile) Parameters in rats.

Parameter	Control	100 mg/kg	200 mg/kg	400 mg/kg
Glucose (mg/dL)	78.11±7.00	72.53±6.88	69.56±8.04	74.30±7.12
Total Bilirubin (mg/dL)	0.206±0.05	0.204±0.06	0.205±0.05	0.200±0.04
Bilirubin direct (mg/dL)	0.1±0.03	0.1±0.03	0.1±0.04	0.1±0.04
Creatinine (mg/dL)	0.92±0.04	0.93±0.05	0.94±0.03	0.95±0.04
BUN (mg/dL)	19.20±1.55	18.58±1.12	18.75±1.56	18.41±1.32
AST (IU/L)	126.1±7.10	124.2±6.05	127.1±5.00	122.4±5.02
ALT (IU/L)	34.10±3.21	32.88±2.79	34.46±2.58	35.96±2.47
ALP (IU/L)	77.05±4.29	68.12±4.00	66.52±4.34	67.10±4.20
Total cholestrol (mg/dL)	59.18±5.44	57.00±5.21	56.75±5.00	57.10±4.92
Total protein (g/dL)	7.99±0.70	7.48±0.58	7.52±0.72	7.64±0.70
Albumin (g/dL)	2.54±0.05	2.70±0.06	2.72±0.04*	2.68±0.04
Urea(mg/dL)	54.48±2.98	56.08±3.88	55.14±2.55	56.45±1.77
Uric acid (mg/dL)	1.94±0.10	1.28±0.15**	1.31±0.14**	1.28±0.12**
Na m.mol	142.64±5.12	141.7±5.06	140.72±4.56	140.21±4.88
K m.mol	21.46±2.37	18.20±1.82	21.20±1.99	20.20±2.54**
Cl m.mol	100.00±5.10	100.44±5.00	98.78±5.42	100.12±5.00
HDL(mg/dL)	13.16±1.45	13.08±1.56	13.43±1.12	13.00±2.88
LDL(mg/dL)	42.22±2.80	42.29±3.08	42.00±3.61	42.20±3.00
VLDL(mg/dl)	16.45±2.00	16.16±2.48	16.36±1.97	15.88±1.56
Triglycerides (mg/dl)	87.52±3.02	86.10±2.09	87.16±3.00	87.47±2.62

Values are mean of 6 animals ± S.E.M. (Dunnet's test). *P<0.05; **P<0.01. Vs Control group

Table-7 Urine Analysis

<i>Parameters</i>	Control	100 mg/kg	200 mg/kg	400 mg/kg
Colour	Yellow	Yellow	Yellow	Yellow
Transparency	Clear	Slightly turbid	Slightly cloudy	Slightly turbid
Specific gravity	1.01	1.01	1.01	1.01
PH	>7.2	>8.0	>8.0	>9.0
Protein	Nil	3+	3+	3+
Glucose	Nil	Nil	Nil	Nil
Bilirubin	-ve	-ve	-ve	-ve
Ketones	-ve	+ve	+ve	+ve
Blood	Absent	Absent	Absent	Absent
Urobilinogen	Normal	Abnormal	Abnormal	Abnormal
Pus cells	0-cells/HPF	1-cell/HPF	2-cells/HPF	1-cell/HPF
RBCs	Nil	Nil	0-1cells/HPF	Nil
Epithelial cells	Nil	1-cell/HPF	Nil	1-cell/HPF
Crystals	Nil	Nil	Nil	Nil
Casts	Nil	Nil	Nil	Nil
Others	Bacteria seen	Bacteria seen	Bacteria seen	Bacteria seen

Values are mean of 6 animals \pm S.E.M. (Dunnett's test). *P<0.05; **P<0.01. vs. *control group*

Table 8. Effect of oral administration of Takkolathi Chooranam on organ weight

Dose (mg/kg)	Control	100 mg/kg	200 mg/kg	400 mg/kg
Heart(g)	0.69 \pm 0.06	0.58 \pm 0.11	0.60 \pm 0.09	0.59 \pm 0.04
Liver(g)	4.52 \pm 0.52	4.19 \pm 0.41	4.92 \pm 0.60	4.82 \pm 0.45
Lung(g)	0.74 \pm 0.04	0.68 \pm 0.06	0.72 \pm 0.05	0.68 \pm 0.1
Spleen(g)	0.64 \pm 0.12	0.69 \pm 0.04	0.71 \pm 0.06	0.66 \pm 0.05
Kidney(g)	1.28 \pm 0.3	1.23 \pm 0.03	1.26 \pm 0.2	1.42 \pm 0.05
Testis(g)	1.02 \pm 0.03	0.98 \pm 0.05	0.92 \pm 0.04	0.94 \pm 0.03
Ovary(g)	0.04 \pm 0.01	0.04 \pm 0.01	0.04 \pm 0.02	0.04 \pm 0.02
Brain	0.79 \pm 0.06	0.76 \pm 0.05	0.77 \pm 0.04	0.75 \pm 0.04
Pancreas	1.44 \pm 0.06	1.35 \pm 0.07	1.38 \pm 0.09	1.42 \pm 0.10
Uterus	0.70 \pm 0.08	0.74 \pm 0.09	0.71 \pm 0.05	0.70 \pm 0.06

Values are mean \pm S.E.M. (Dunnett's t' test). ^{ns}P > 0.05; Vs Control N=6.

PHARMACOLOGICAL STUDY

Table-1: Effect of Takkolathi Chooranam on isolated Guinea pig ileum preparation

Sl. No	Dose of Histamine (µg/ml)	Percent of maximum response	
		Histamine alone	Histamine+Takkolathi Chooranam (% of Inhibition)
1	10	1.85±0.17	1.12±0.12 (38.88%)*
2	20	2.21±0.21	1.56±0.63 (31.81%)*
3	40	2.86±0.32	1.70±0.44 (40.55%)*

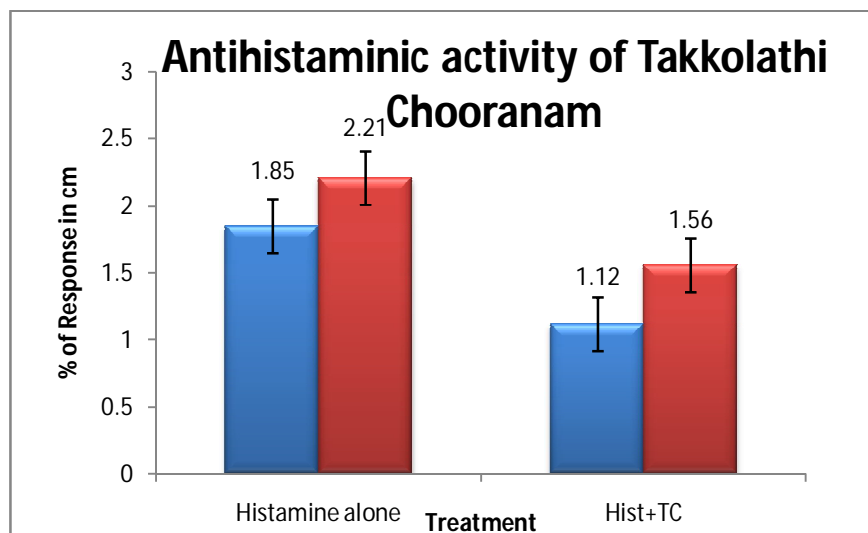
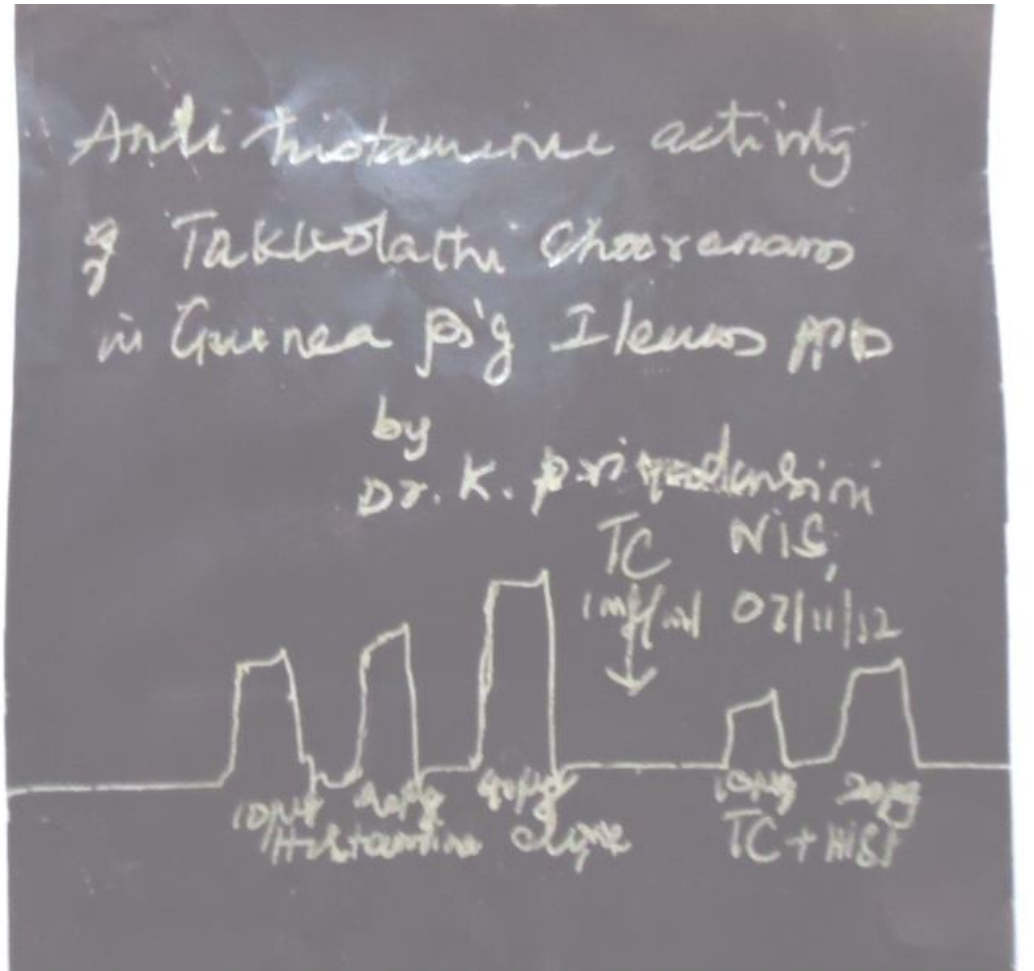
Values are expressed in mean ± SEM, *p< 0.01 compared with histamine induced contraction (28mm as 100%); n=3.

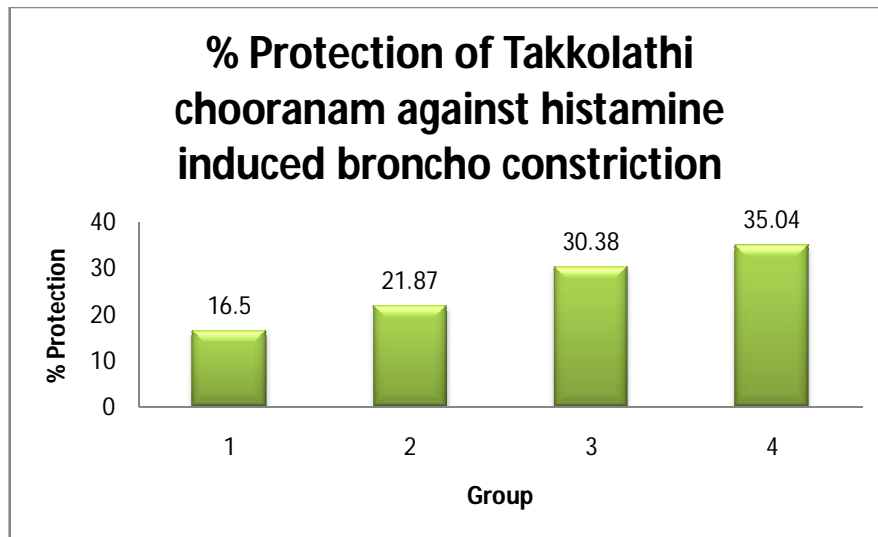
Table-2: Bronchodilator effect of Takkolathi chooranam on Histamine induced Bronchoconstriction.

Treatment	Pre-Treatment Exposition in seconds	Post-Treatment Exposition in seconds	% Protection
Takkolathi Chooranam 100mg/kg. p.o.	94.27 ±3.48	112.91±4.11	16.50%
Takkolathi Chooranam 200mg/kg. p.o.	102.56±3.82	131.28±4.76**	21.87%
Takkolathi Chooranam 400mg/kg. p.o.	97.21±4.00	139.63±5.02**	30.38%
Promethazine (300mg/kg, p.o)	105.50±4.33	162.42±5.12**	35.04%

N=6; Values are expressed as mean ± SEM; *Significant between pre and post treatment time (Student's - 't') **P<0.01.

ANTI HISTAMINE ACTIVITY OF TAKKOLATHI CHOORANAM IN GUINEA
PIG:





STATISTICAL ANALYSIS:

Paired t test for Symptoms before and after treatment for Eraippu patients:

Variable	Obs	Mean	Std.dev	t.value	P value
BTSym	20	4.1	1.071	13.21	P<0.0001
ATSym	20	1.15	1.424		

Symptoms before treatment is 4.10 and after treatment is 1.15 which is statistically significant($p < 0.0001$).

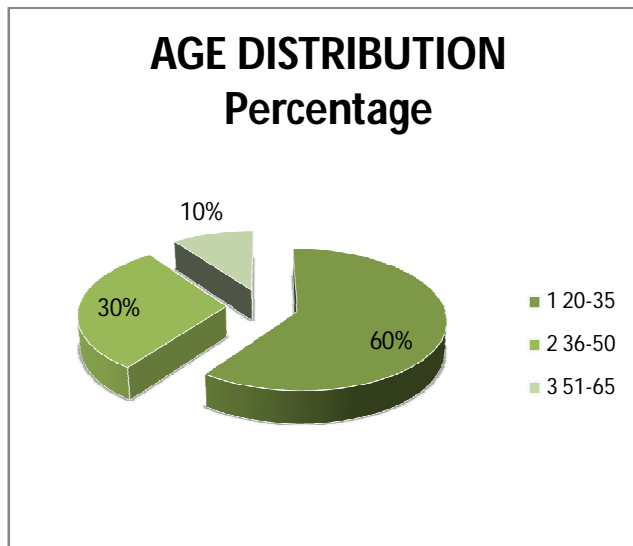
Paired t test for PEFR before and after treatment for Eraippu patients:

Variable	Obs	Mean	Std.dev	t.value	P value
BT PEFR	20	183.5	21.831	-8.73	P<0.0001
AT PEFR	20	226.5	26.413		

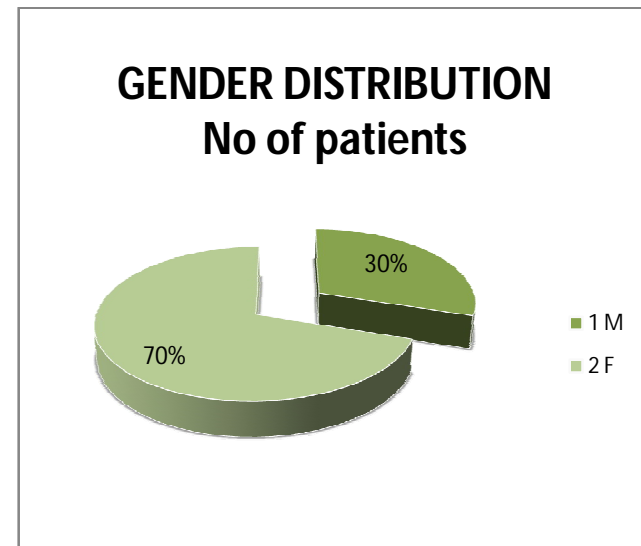
PEFR before treatment is 183.50 and after treatment is 226.50 which is statistically significant($p < 0.0001$).

CLINICAL STUDY

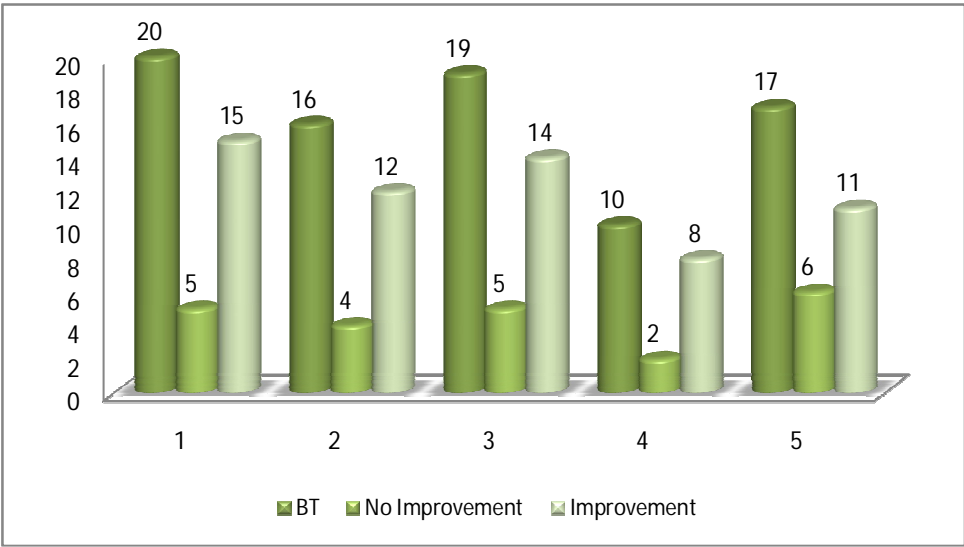
AGE DISTRIBUTION		
SL No	Age (years)	Percentage
1	20-35	60
2	36-50	30
3	51-65	10



GENDER DISTRIBUTION			
SL No	Gender	No of patients	Percentage
1	M	6	30
2	F	14	70



IMPROVEMENT SHOWING SIGNS & SYMPTOMS BEFORE AND AFTER TREATMENT OF ERAIPPU PATIENTS					
SL No	Symptoms	No of patients with symptoms			Improvement percentage
		BT	AT		
			No Improvement	Improvement	
1	Wheeze	20	5	15	75
2	Dyspnoea	16	4	12	75
3	Cough	19	5	14	74
4	Chest tightness	10	2	8	80
5	Expectoration	17	6	11	65



PROGNOSIS IN SIGNS AND SYMPTOMS OF BRONCHIAL ASTHMA												
SL NO	OP/IP No	Name	Wheeze		Dyspnoea		Cough		Chest Tightness		Expectoration	
			BT	AT	BT	AT	BT	AT	BT	AT	BT	AT
1	3951	Mrs.G.Dhanaselvi	+	-	+	-	+	+	+	-	+	-
2	4241	Miss.Chitra	+	-	+	+	+	+	+	+	+	+
3	4263	Mrs.Muniyammal	+	-	-	-	+	-	-	-	+	-
4	4271	Mrs.Esweri	+	+	+	-	+	-	+	-	+	+
5	C82597	Mr.R.Ranjit	+	-	+	-	+	-	-	-	+	-
6	C83283	Mrs.Sujatha	+	-	+	-	+	+	+	-	+	+
7	C83365	Mrs.G.Revathy	+	-	-	-	+	-	-	-	+	-
8	C83360	Mr.Bharathi Mohan	+	-	+	-	+	-	-	-	+	-
9	C80148	Mrs.B.Komala	+	-	+	-	+	-	+	-	+	+
10	C78368	Mrs.Dhanalakshmi	+	-	+	-	+	-	-	-	+	-
11	C84545	Mr.P.Hariharan	+	-	-	-	+	-	-	-	-	-
12	C86210	Mrs.B.Sirumbai	+	+	+	+	+	-	+	-	+	-
13	C85376	Mrs.Amuthavalli	+	+	+	-	+	+	+	+	+	-
14	C85322	Mrs.K.Panchatcharam	+	+	+	-	+	-	+	-	+	+
15	C93190	Mrs.S.Venda	+	-	+	-	+	-	-	-	+	-
16	C89071	Mr.M.Muralidharan	+	-	+	-	-	-	-	-	-	-
17	C85704	Mrs.A.R.Anandhi	+	+	+	+	+	+	+	-	+	+
18	C85145	Mr.Selvapalani	+	-	-	-	+	-	-	-	+	-
19	C88370	Miss.Asmathnisha	+	-	+	-	+	-	-	-	-	-
20	C58877	Mr.Sathish Kumar	+	-	+	+	+	-	+	-	+	-

LAB INVESTINGATIONS – BEFORE TREATMENT BRONCHIAL ASTHMA PATIENTS

SL No	OP/IP No	Age/Sex	Hb	TC	Dc (in %)				ESR (mm/hr)		BS (mg/dl)		AEC	Urea	Creat	OT	PT	Al.P	T.Chol	Urine			
					P	L	E	M	1/2	1	F	PP/R								Alb	Sug	Dep	
																						Pus cells	Epi cells
1	3951	34/F	11.2	12000	70	22	6	2	6	14	80	130	450	20	0.7	25	22	168	170	Nil	Nil	1-2	1-2
2	4241	28/F	12.3	10000	60	28	12	0	6	12	92	120	420	28	0.9	33	30	130	194	Nil	Nil	1-2	1-2
3	4263	35/F	10.5	10500	70	23	7	0	4	8	100	130	350	20	0.7	25	22	96	210	Nil	Nil	1-2	2-3
4	4271	42/F	11.8	11000	66	26	8	0	6	12	98	126	630	16	0.8	21	18	130	206	Nil	Nil	1-2	2-3
5	C82597	33/M	10.8	11000	71	21	8	0	6	10	102	131	500	21	0.7	26	23	120	214	Nil	Nil	2-3	1-2
6	C83283	42/F	14.3	8000	67	25	6	2	8	14	104	127	180	17	0.6	22	19	168	218	Nil	Nil	1-2	1-2
7	C83365	47/F	13.8	8500	64	29	6	1	40	16	99	124	266	28	0.9	33	30	192	208	Nil	Nil	1-2	1-2
8	C83360	25/M	13.9	11300	74	15	11	0	6	18	101	134	412	24	0.8	29	26	216	212	Nil	Nil	2-3	1-2
9	C80148	30/F	13	9000	72	22	6	0	8	10	100	132	444	22	0.7	27	24	120	210	Nil	Nil	1-2	2-3
10	C78368	48/F	10	9400	70	23	5	2	6	10	98	130	233	20	0.7	25	22	120	206	Nil	Nil	1-2	1-2
11	C84545	39/M	14	7000	69	23	8	0	12	18	103	129	560	19	0.6	24	21	216	216	Nil	Nil	2-3	2-3
12	C86210	58/F	10.8	6800	63	31	6	0	8	20	106	123	388	32	1.1	37	34	240	222	Nil	Nil	2-3	1-2
13	C85376	25/F	12.8	10200	66	19	14	1	12	18	110	126	777	24	0.8	29	26	216	210	Nil	Nil	1-2	1-2
14	C85322	54/F	13	9700	69	19	12	0	12	16	104	129	540	19	0.6	24	21	192	218	Nil	Nil	1-2	1-2
15	C93190	25/F	12.5	6900	73	20	6	1	8	12	98	133	460	23	0.8	28	25	144	206	Nil	Nil	1-2	2-3
16	C89071	30/M	14.2	7800	70	25	5	0	6	10	96	130	356	20	0.7	25	22	120	202	Nil	Nil	2-3	1-2
17	C85704	31/F	9.8	8200	64	31	5	0	8	16	84	124	288	35	1.2	40	37	192	178	Nil	Nil	1-2	1-2
18	C85145	39/M	13.8	9200	62	20	15	3	8	10	110	122	420	22	0.7	27	24	120	203	Nil	Nil	1-2	1-2
19	C88370	20/F	10.2	10400	68	27	5	0	6	12	108	128	322	18	0.6	23	20	144	226	Nil	Nil	2-3	2-3
20	C58877	24/M	14	6800	70	21	8	1	8	12	104	130	350	20	0.7	25	22	123	218	Nil	Nil	1-2	1-2

LAB INVESTINGATIONS – AFTER TREATMENT BRONCHIAL ASTHMA PATIENTS																							
SL No	OP/IP No	Age/Sex	Hb	TC	Dc (in %)				ESR (mm/hr)		BS (mg/dl)		AEC	Urea	Creat	OT	PT	Al.P	T.Chol	Urine			
					P	L	E	M	1/2	1	F	PP/R								Alb	Sug	Dep	
1	3951	34/F	11.4	12000	74	22	3	1	6	12	81	137	165	20	0.7	25	24	171	172	Nil	Nil	1-2	1-2
2	4241	28/F	12.5	10000	66	28	6	0	4	10	90	129	244	28	0.9	21	32	133	196	Nil	Nil	1-2	1-2
3	4263	35/F	10.7	10500	70	23	5	2	6	4	92	133	240	20	0.7	22	24	99	212	Nil	Nil	1-2	2-3
4	4271	42/F	12	11000	70	26	4	0	3	10	92	133	420	16	0.8	25	20	133	208	Nil	Nil	1-2	2-3
5	C82597	33/M	11	11000	74	21	5	0	2	12	92	137	350	21	0.7	24	25	123	216	Nil	Nil	2-3	1-2
6	C83283	42/F	14.5	8000	72	25	3	0	6	12	95	135	150	17	0.6	25	21	171	220	Nil	Nil	1-2	1-2
7	C83365	47/F	14	8500	63	29	6	2	8	12	94	126	270	28	0.9	26	32	195	210	Nil	Nil	1-2	1-2
8	C83360	25/M	14.1	11300	78	15	6	1	4	14	88	141	310	24	0.8	27	28	219	214	Nil	Nil	2-3	1-2
9	C80148	30/F	13.2	9000	72	22	5	1	8	12	91	135	222	22	0.7	29	26	123	212	Nil	Nil	1-2	2-3
10	C78368	48/F	10.2	9400	69	23	6	2	6	8	91	132	230	20	0.7	29	24	123	208	Nil	Nil	1-2	1-2
11	C84545	39/M	14.2	7000	71	23	6	0	8	12	93	134	300	19	0.6	33	23	219	218	Nil	Nil	2-3	2-3
12	C86210	58/F	11	6800	63	31	4	2	4	16	99	126	190	32	1.1	24	36	243	224	Nil	Nil	2-3	1-2
13	C85376	25/F	13	10200	70	19	10	1	6	12	95	133	400	24	0.8	25	28	219	212	Nil	Nil	1-2	1-2
14	C85322	54/F	13.2	9700	72	19	8	1	8	12	92	135	269	19	0.6	28	23	195	220	Nil	Nil	1-2	1-2
15	C93190	25/F	12.7	6900	77	20	2	1	4	8	89	140	193	23	0.8	33	27	147	208	Nil	Nil	1-2	2-3
16	C89071	30/M	14.4	7800	69	25	5	1	4	6	91	132	290	20	0.7	37	24	123	204	Nil	Nil	2-3	1-2
17	C85704	31/F	10	8200	63	31	4	2	2	4	88	126	189	35	1.2	40	39	195	180	Nil	Nil	1-2	1-2
18	C85145	39/M	14	9200	72	20	8	0	4	8	95	135	280	22	0.7	27	26	123	205	Nil	Nil	1-2	1-2
19	C88370	20/F	10.4	10400	68	27	5	0	2	10	98	131	166	18	0.6	23	22	147	228	Nil	Nil	2-3	2-3
20	C58877	24/M	14.2	6800	73	21	5	1	4	12	93	136	211	20	0.7	25	24	126	220	Nil	Nil	1-2	1-2

LAB INVESTIGATIONS – PEFR BRONCHIAL ASTHMA PATIENTS					
SL No	OP/IP No	Name	Age/Sex	Peak Expiratory Flow Rate	
				Before treatment	After treatment
1	3951	Mrs.G.Dhanaselvi	34/F	150	180
2	4241	Miss.Chitra	28/F	160	210
3	4263	Mrs.Muniyammal	35/F	170	220
4	4271	Mrs.Esweri	42/F	170	230
5	C82597	Mr.R.Ranjit	33/M	180	180
6	C83283	Mrs.Sujatha	42/F	170	210
7	C83365	Mrs.G.Revathy	47/F	190	220
8	C83360	Mr.Bharathi Mohan	25/M	200	250
9	C80148	Mrs.B.Komala	30/F	180	230
10	C78368	Mrs.Dhanalakshmi	48/F	230	250
11	C84545	Mr.P.Hariharan	39/M	190	260
12	C86210	Mrs.B.Sirumbai	58/F	170	220
13	C85376	Mrs.Amuthavalli	25/F	200	200
14	C85322	Mrs.K.Panchatcharam	54/F	160	220
15	C93190	Mrs.S.Venda	25/F	190	270
16	C89071	Mr.M.Muralidharan	30/M	200	280
17	C85704	Mrs.A.R.Anandhi	31/F	180	220
18	C85145	Mr.Selvapalani	39/M	220	230
19	C88370	Miss.Asmathnisha	20/F	160	210
20	C58877	Mr.Sathish Kumar	24/M	220	240

PHYSICAL PROPERTIES: (Table B)

S.NO	Characteristic test	Results
1	pH	3.90
2	Ash Value	0.06
3	Water soluble ash	0.03
4	Acid insoluble ash	0.03

QUALITATIVE ANALYSIS:

S.NO	PARAMETERS	RESULTS
1	Calcium	Present
2	Sulphate	Present
3	Sodium	Present
4	Iron	Present
5	Chloride	Absent
6	Starch	Present
7	Flavonoids	Absent
8	Aminoacids	Absent
9	Tannic acid	Present
10	Glucose	Absent

Table C:**ATOMIC ABSORPTION SPECTROPHOTOMETER****METAL CONTENT:**

SAMPLE NAME	Fe (ppm)	Zn (ppm)	K(ppm)	Na (ppm)	Ca (ppm)	Mg (ppm)
KUMARI PARPAM	0.13	-	-	-	-	-

Table 1: Dose finding experiment and its behavioral Signs of Toxicity

No	Dose mg/kg	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
1	500	+	-	-	+	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-
2	1000	+	-	-	+	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-
3	2000	+	-	-	+	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-

1. Alertness 2. Aggressiveness 3. Pile erection 4. Grooming 5. Gripping 6. Touch Response
 7. Decreased Motor Activity 8. Tremors 9. Convulsions 10. Muscle Spasm 11. Catatonia 12.
 Muscle relaxant 13. Hypnosis 14. Analgesia 15. Lacrimation 16. Exophthalmos 17. Diarrhoea
 18. Writhing 19. Respiration 20. Mortality

Table 2. Body wt (g) of rats exposed to *Kumari Parpam* for 28days.

Dose (mg/kg/day)	Days				
	1	7	14	21	28
Control	112.54±5.05	110.56±6.00	114.82±5.25	118.05±8.11	123.00±5.69
50	117.07±6.31	119.21±5.15	122.19±5.76	125.64±10.00	126.56±6.08
100	114.26±5.44	118.13±6.88	121.08±6.15	124.18±7.14	127.37±8.14
200	112.13±7.27	115.62±5.34	118.12±6.98	121.02±6.33	124.51±7.32

Values are mean ± S.E.M. (Dunnet't' test). ^{ns}P > 0.05; Vs Control N=6.

Table 3. Food (g/day) intake of rats exposed to *Kumari Parpam* for 28days.

Dose (mg/kg/day)	Days (gms/rats)				
	1	7	14	21	28
Control	47.22±2.45	48.12±2.17	46.10±2.28	47.61±2.44	47.85±3.22
50	45.20±2.62	45.75±2.40	46.24±2.45	47.45±2.96	48.10±3.46
100	42.34±2.12	42.40±2.47	44.45±2.34	45.84±2.10	46.45±3.21
200	42.52±2.25	44.11±2.84	46.47±2.54	45.33±2.12	45.66±3.00

Values are mean ± S.E.M. (Dunnet't' test). ^{ns}P > 0.05; Vs Control N=6.

Table 4. Water (ml/day) intake of rats exposed to *Kumari Parpam* for 28days.

	Days(ml/rat)				
Dose (mg/kg/day)	1	7	14	21	28
Control	51.24±2.75	52.12±2.86	55.28±3.18	58.37±3.18	45.11±3.33
50	50.42±2.58	50.86±3.02	45.23±4.00	42.21±3.42**	40.49±2.48
100	48.98±2.96	49.58±3.78	40.87±3.39*	47.12±3.12	40.17±3.24
200	52.44±3.12	54.00±3.12	55.32±3.00	45.25±4.00*	42.16±3.00

Values are mean ± S.E.M. (Dunnet 't' test). *P<0.05; **P<0.01. Vs Control N=6.

Table 5. Hematological parameters after 28days treatment with *Kumari Parpam* in rats.

Parameter	Control	50 mg/kg	100 mg/kg	200 mg/kg
RBC (millions/cu.mm)	5.13±0.41	5.15±0.41	5.26±0.32	5.32±0.41
Hb (g/dl)	13.01±0.28	13.83±0.98	14.0±1.1	14.12±1.0
PCV (%)	42.11±1.05	44.5±4.1	44.0±2.5	45.15±1.2
WBC(cells/cu.mm)	7368±434.12	8045±337.2	8428±415.7	8643±452.3
Neutrophil (%)	54.45±4.58	42.24 ±2.7	47.22±3.0	47.10±3.2
Lymphocytes (%)	38.01±1.40	51.2±3.03*	47.31±3.4	46.18±3.2
Eosinophil's (%)	7.2±0.41	5.1±0.75*	5.3±0.41*	5.10±0.43*
Monocytes (%)	4.0±0.3	3.2±0.4	3.0±0.3	2.4±0.2**
Basophils (%)	0±0	0±0	0±0	0±0
Platelets (10 ⁵ cells/cu.mm)	1.41±0.05	1.80±0.07*	1.82±0.06*	2.0±0.19**
MCV(fl)	76.1±2.2	82.6±2.0	81.5±2.2	83.2±5.2
MCHC (pg)	26.2±1.8	25.82±1.5	28.24±1.6	30.00±2.0

Values are mean ± S.E.M. (Dunnet 't' test). *P<0.05; **P<0.01. Vs Control N=6.

**Table 6. Effect of treatment with *Kumari Parpam* biochemical (LFT, RFT, Lipid Profile)
Parameters in rats.**

Parameter	Control	50 mg/kg	100 mg/kg	200 mg/kg
Glucose (mg/dL)	72.42±6.20	68.42±7.32	66.32±7.13	67.42±7.28
Total Bilirubin(mg/dL)	0.205±0.05	0.214±0.06	0.212±0.05	0.216±0.04
Bilirubin direct (mg/dL)	0.1±0.04	0.1±0.05	0.1±0.04	0.1±0.05
Creatinine (mg/dL)	0.90±0.06	0.89±0.05	0.98±0.03*	0.98±0.03*
BUN (mg/dL)	19.22±1.68	18.50±1.19	17.83±1.80	18.20±1.61
AST (IU/L)	134.8±6.24	121.3±6.33	119.3±6.12	122.0±4.07
ALT (IU/L)	38.22±3.50	32.18±2.60	30.14±2.56	31.62±2.28
ALP (IU/L)	79.30±4.32	72.65±4.32	66.32±4.36	65.40±4.15
Total cholestrol (mg/dL)	55.81±5.78	57.61±5.68	55.84±5.21	59.61±4.88
Total protein (g/dL)	8.42±0.27	7.90±0.23	7.53±0.75	7.46±0.72
Albumin (g/dL)	2.68±0.08	2.72±0.05	2.74±0.06	2.72±0.05
Urea(mg/dL)	55.24±1.53	54.24±3.64	62.12±2.15**	53.80±1.65
Uric acid (mg/dL)	1.6±0.12	1.6±0.18	2.6±0.14**	2.2±0.18**
Na m.mol	140.82±5.20	141.5±5.04	142.14±5.12	141.16±5.12
K m.mol	20.20±2.88	19.40±1.42	20.0±1.48	20.10±2.24
Cl m.mol	100.05±4.24	102.20±5.21	98.80±4.72	101.24±4.16
HDL(mg/dL)	13.02±1.44	13.20±1.70	13.02±1.45	13.20±2.12
LDL(mg/dL)	42.00±2.83	44.15±3.56	42.31±3.92	43.20±3.34
VLDL(mg/dl)	16.38±2.62	15.82±2.34	16.00±1.64	15.06±1.28
Triglycerides (mg/dl)	87.24±3.00	85.14±2.20	86.28±3.24	88.40±2.70

Values are mean ± S.E.M. (Dunnet 't' test). *P<0.05; **P<0.01. Vs Control

Table-7 Urine Analysis

<i>Parameters</i>	Control	50 mg/kg	100 mg/kg	200 mg/kg
Colour	Yellow	Yellow	Yellow	Yellow
Transparency	Clear	Slightly turbid	Slightly cloudy	Slightly turbid
Specific gravity	1.01	1.01	1.01	1.01
PH	>7.2	>8.0	>8.0	>9.0
Protein	Nil	3+	3+	3+
Glucose	Nil	Nil	Nil	Nil
Bilirubin	-ve	-ve	-ve	-ve
Ketones	-ve	+ve	+ve	+ve
Blood	Absent	Absent	Absent	Absent
Urobilinogen	Normal	Abnormal	Abnormal	Abnormal
Pus cells	0-cells/HPF	1-cell/HPF	2-cells/HPF	1-cell/HPF
RBCs	Nil	Nil	0-1 cells/HPF	Nil
Epithelial cells	Nil	1-cell/HPF	Nil	1-cell/HPF
Crystals	Nil	Nil	Nil	Nil
Casts	Nil	Nil	Nil	Nil
Others	Bacteria seen	Bacteria seen	Bacteria seen	Bacteria seen

Table 8. Effect of *Kumari Parpam* on organ weight

Dose (mg/kg)	Control	50 mg/kg	100 mg/kg	200 mg/kg
Heart(g)	0.67±0.09	0.57±0.12	0.61±0.08	0.58±0.05
Liver(g)	4.50±0.50	4.17±0.44	4.90±0.60	4.80±0.42
Lung(g)	0.7±0.05	0.65±0.05	0.70±0.05	0.67±0.04
Spleen(g)	0.64±0.05	0.67±0.04	0.70±0.06	0.68±0.05
Kidney(g)	1.02±0.03	1.04±0.04	1.16±0.4	1.22±0.05
Testis(g)	1.00±0.03	0.94±0.05	0.93±0.04	0.95±0.03
Ovary(g)	0.04±0.02	0.04±0.01	0.04±0.02	0.04±0.02
Brain	0.77±0.06	0.67±0.04	0.69±0.03	0.72±0.05
Pancreas	1.42±0.07	1.36±0.08	1.41±0.11	1.61±0.10
Uterus	0.69±0.07	0.84±0.08	0.72±0.06	0.72±0.07

Values are mean ± S.E.M. (Dunnet't test). ^{ns}P > 0.05; Vs Control N=6.

Pharmacological study:

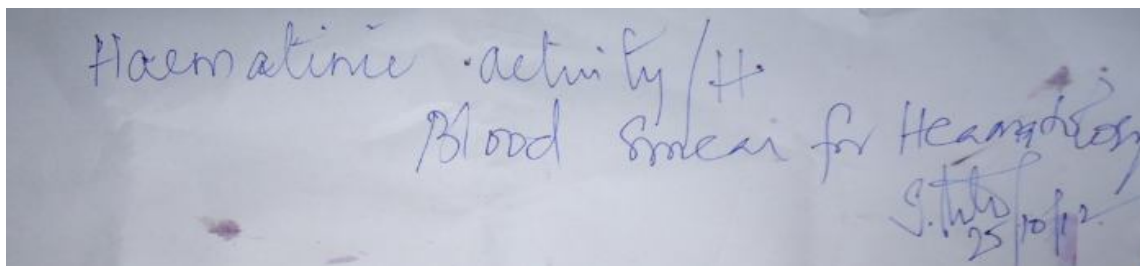


Table 1: Effect of Phenylhydrazine (10mg/kg, p.o. daily for 7 days) alone on Hematological parameters.

Parameters	Group 1 (Normal)	Group 2 (Anemic)	Group 3 (Anemic)	Group 4 (Anemic)	Group 5 (Anemic)	Group 6 (Anemic)
Hb (g/dl)	18.24 ± 0.52	13.4±0.30**	13.68±0.28**	13.22±0.42**	12.98±0.30**	12.64 ± 0.32**
PCV (%)	54.10 ±1.73	40.54 ± 2.48**	41.50 ± 1.68**	41.34 ± 2.44**	40.51 ± 2.12**	40.34 ± 2.87**
RBC (x10⁶/ml)	6.53 ± 0.19	4.28 ± 0.32**	4.10 ± 0.22**	4.85 ± 0.25**	4.76 ± 0.24**	4.22 ± 0.39**
MCV (fl)	72.70 ± 2.88	83.24±4.2	87.45±7.00	87.16±3.10	86.98±7.33	90.01± 3.40
MCH (pg)	23.81 ± 1.69	29.36±1.44	29.45±1.75	30.15±1.23*	30.17±0.92*	30.12 ± 2.54*
MCHC (g/dl)	34.12 ± 0.48	33.17±0.6	33.32±0.85	34.00±1.62	34.30±3.22	30.77 ± 1.41

Values are mean ± S.E.M. (Dunnet't' test). *P<0.05; **P<0.01 Vs Control N=6.

Table 2: Hematological parameter of rats after Seven days treatment with Kumari Parpam.

Parameters	Group 1 (Normal)	Group 2 (Anemic Control)	Group 3 (50 mg/kg)	Group 4 (100 mg/kg)	Group 5 (200 mg/kg)	Group 6 (Heamatinic syrup)
Hb (g/dl)	19.33 ± 1.23*	14.12 ± 1.02	16.12 ± 0.55	16.98 ± 1.00	17.00 ± 0.86	21.46 ± 2.00**
PCV (%)	55.26 ± 1.48*	44.72 ± 2.4	45.12 ± 2.36	47.10 ± 2.64	47.10 ± 3.22	57.02 ± 2.41**
RBC (x10 ⁶ /ml)	7.35 ± 0.20*	5.10 ± 0.24	6.88 ± 0.20	7.12 ± 0.13*	8.02 ± 0.17**	8.11 ± 1.10**
MCV (fl)	79.12 ± 2.41	78.5 ± 2.6	81.0 ± 1.4	78.1 ± 2.8	76.16 ± 5.6	74.45 ± 2.13
MCH (pg)	24.94 ± 1.56	27.3 ± 1.6	24.91 ± 1.1	29.6 ± 1.3	30.15 ± 2.3	22.15 ± 1.22
MCHC (g/dl)	33.17 ± 1.34	30.48 ± 1.22	26.10 ± 1.4	25.12 ± 1.0	28.5 ± 1.0	31.64 ± 2.4

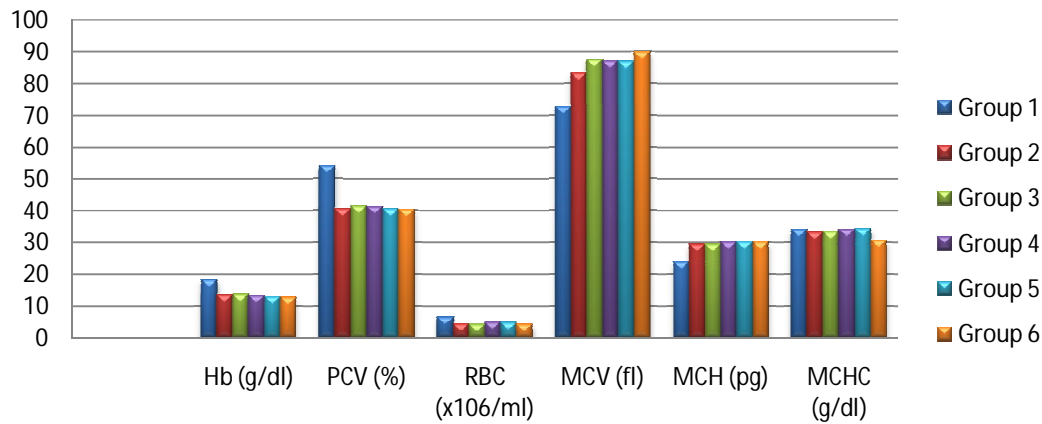
Values are mean ± S.E.M. (Dunnet't' test). *P<0.05; **P<0.01 Vs Control N=6.

Table 3: Hematological parameters of rats after 14 days treatment with Kumari Parpam.

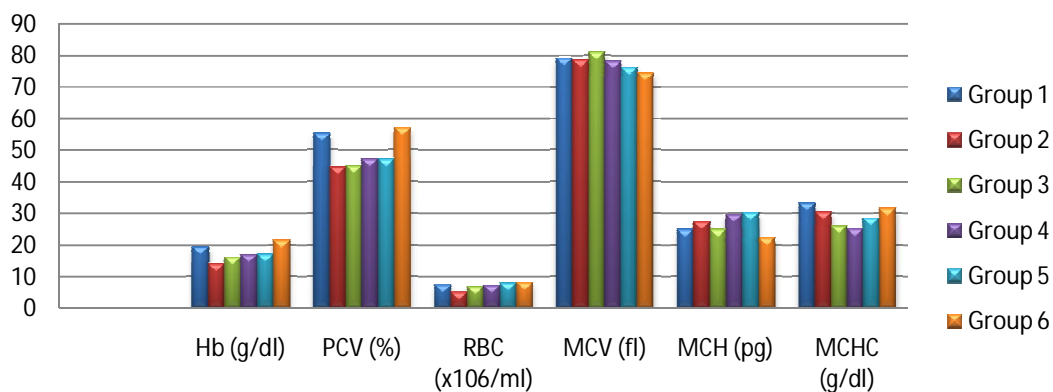
Parameters	Group 1 (Normal)	Group 2 (Anemic Control)	Group 3 (50 mg/kg)	Group 4 (100 mg/kg)	Group 5 (200 mg/kg)	Group 6 (Heamatinic syrup)
Hb (g/dl)	18.33 ± 1.41**	10.15 ± 1.10	20.65 ± 0.90**	20.44 ± 0.85**	20.12 ± 1.65**	21.58 ± 1.50**
PCV (%)	46.44 ± 1.24	43.20 ± 1.12	42.10 ± 3.8	42.34 ± 2.0	44.11 ± 1.4	57.37 ± 1.12
RBC (x10 ⁶ /ml)	4.92 ± 0.30	4.90 ± 0.34	4.78 ± 0.32	4.67 ± 0.25	4.70 ± 0.24	5.54 ± 0.27
MCV (fl)	75.44 ± 2.15**	92.88 ± 2.49	90.32 ± 2.01	77.45 ± 1.50**	80.20 ± 2.48**	75.41 ± 2.45**
MCH (pg)	27.10 ± 2.56	34.00 ± 2.01	30.12 ± 1.88	26.71 ± 1.50	27.00 ± 1.46	28.30 ± 2.79
MCHC (g/dl)	32.02 ± 2.61	33.77 ± 1.02	33.23 ± 1.46	33.65 ± 0.77	33.10 ± 2.34	33.51 ± 2.18

Values are mean ± S.E.M. (Dunnet't' test). **P<0.01 Vs Control N=6.

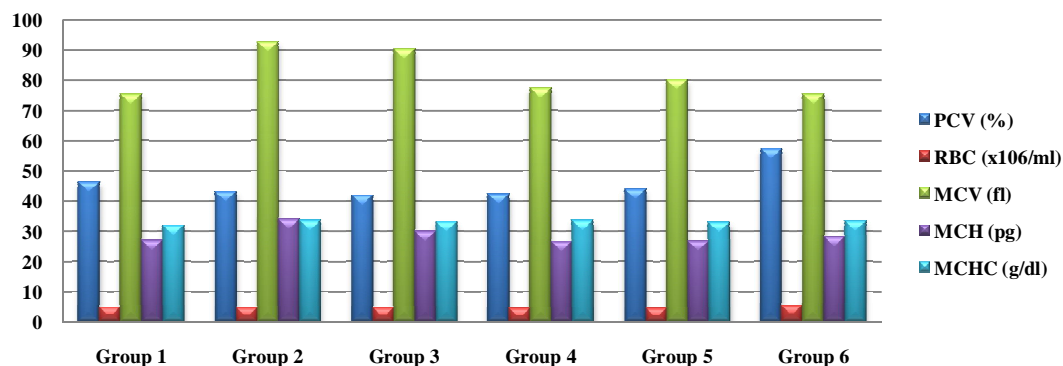
Effect of Phenylhydrazine alone on Hematological parameters



Hematological parameter of rats after Seven days treatment with Kumari Parpam



Hematological parameters of rats after 14 days treatment with Kumari Parpam



STATISTICAL ANALYSIS:

Paired t test for Symptoms before and after treatment for Pandu patient:

Variable	Obs	Mean	Std.dev	t.value	P value
BTSym	20	5.45	1.432	9.054	P<0.0001
ATSym	20	2.8	1.609		

Symptoms before treatment is 5.45 and after treatment is 2.80 which is statistically significant ($p<0.0001$).

Paired t test for Hb before and after treatment for Pandu patient:

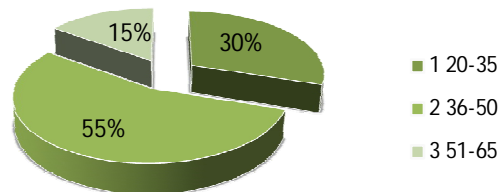
Variable	Obs	Mean	Std.dev	t.value	P value
BT Hb	20	8.58	0.9595	-16.6	P<0.0001
AT Hb	20	9.515	9.515		

Hb before treatment is 8.580 and after treatment is 9.515 which is statistically significant ($p<0.0001$).

CLINICAL STUDY

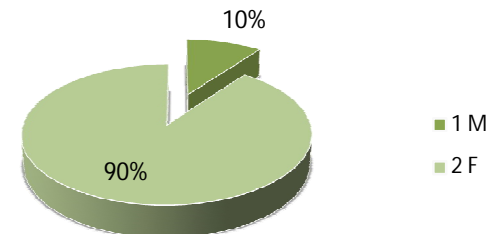
AGE DISTRIBUTION		
SL No	Age (years)	Percentage
1	20-35	30
2	36-50	55
3	51-65	15

AGE DISTRIBUTION
Percentage

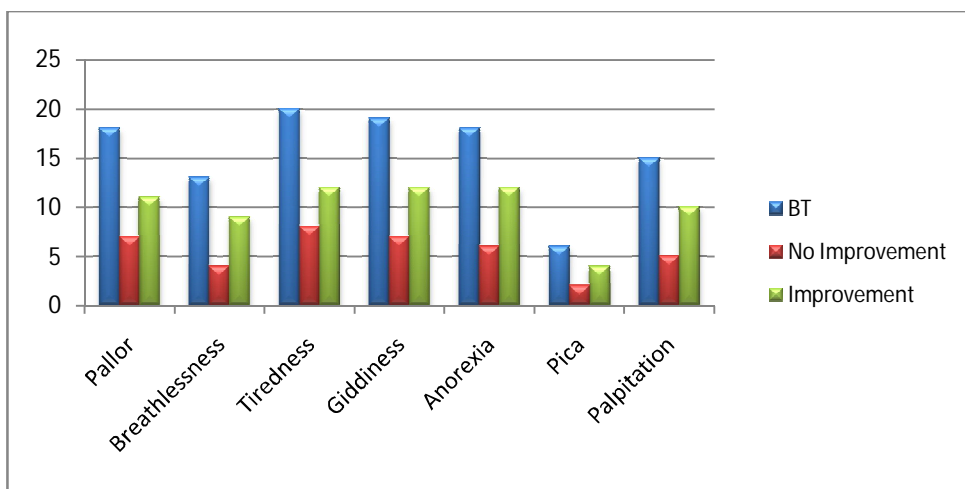


GENDER DISTRIBUTION			
SL No	Gender	No of Patients	Percentage
1	M	2	10
2	F	18	90

GENDER DISTRIBUTION
No of Patients



IMPROVEMENT SHOWING SIGNS & SYMPTOMS BEFORE AND AFTER TREATMENT OF PAANDU PATIENTS					
SL No	Symptoms	No of patients with symptoms			Improvement percentage
		BT	AT		
			No Improvement	Improvement	
1	Pallor	18	7	11	61
2	Breathlessness	13	4	9	69
3	Tiredness	20	8	12	60
4	Giddiness	19	7	12	63
5	Anorexia	18	6	12	66.7
6	Pica	6	2	4	66.7
7	Palpitation	15	5	10	66.7



PROGNOSIS IN SIGNS AND SYMPTOMS OF ANAEMIA																
SL NO	OP No	Name	Pallor		Breathlessness		Tiredness		Giddiness		Anorexia		Pica		Palpitation	
			BT	AT	BT	AT	BT	AT	BT	AT	BT	AT	BT	AT	BT	AT
1	C83474	Mrs.V.Padma	+	+	+	+	+	-	+	-	+	+	+	+	+	+
2	C85480	Mrs.R.Arputham	+	-	-	-	+	-	+	-	+	-	-	-	+	-
3	C85924	Mrs.Dhatchayini	+	-	-	-	+	+	+	+	+	-	-	-	+	+
4	C098088	Mrs.C.Athilakshmi	+	-	+	-	+	-	-	-	+	-	-	-	-	-
5	C99770	Miss.S.Sudha	+	+	+	-	+	+	+	-	+	+	+	+	+	-
6	C97890	Mrs.P.Saratha	+	-	+	+	+	+	+	+	+	-	+	-	+	-
7	C97884	Mr.M.Ravi	+	+	+	-	+	+	+	-	+	+	-	-	+	-
8	C89948	Mrs.R.Panjali	-	-	-	-	+	-	+	-	-	-	+	+	-	-
9	C90117	Mrs.K.G.Govindammal	+	+	+	-	+	-	+	+	+	-	-	-	-	-
10	D00452	Mr.P.Loganathan	+	-	-	-	+	+	+	-	+	-	-	-	+	+
11	C98655	Mrs.K.S.Rani	-	-	-	-	+	-	+	+	-	-	-	-	-	-
12	C91131	Mrs.S.Mythili	+	-	+	-	+	-	+	+	+	+	-	-	+	+
13	C9941	Mrs.Lally Baskaran	+	+	+	+	+	-	+	-	+	-	+	+	+	-
14	C00347	Mrs.I.Banu	+	-	+	-	+	-	+	-	+	-	-	-	+	+
15	C95755	Mrs.S.Maruthi	+	+	+	+	+	+	+	+	+	-	+	-	+	-
16	C67253	Mrs.R.Malliga	+	-	-	-	+	-	+	-	+	-	-	-	+	-
17	C002479	Mrs.K.Gomathi	+	-	-	-	+	-	+	-	+	-	-	-	-	-
18	C90388	Mrs.A.Shanthi	+	-	+	-	+	+	+	+	+	-	-	-	+	-
19	C001189	Mrs.L.Shoba	+	-	+	-	+	-	+	-	+	-	-	-	+	+
20	C97754	Mrs.M.Muthulakshmi	+	+	+	+	+	+	+	+	+	-	-	-	+	-

LAB INVESTINGATIONS – BEFORE TREATMENT ANAEMIA PATIENTS																					
SL No	OP No	Age/Sex	TC	Dc (in %)				ESR (mm/hr)		BS (mg/dl)		Urea	Creat	OT	PT	Al.P	T.Chol	Urine			
				P	L	E	M	1/2	1	F	PP/R							Alb	Sug	Dep	
																				Pus cells	Epi cells
1	C83474	50/F	7500	60	39	1	0	16	32	99	129	26	0.6	29	26	234	193	NIL	NIL	1-2	2-3
2	C85480	60/F	8200	66	31	2	1	12	16	91	121	27	0.5	30	27	188	153	NIL	NIL	1-2	1-2
3	C85924	42/F	9200	71	28	1	0	14	20	88	118	26	0.8	29	28	168	158	NIL	NIL	2-3	2-3
4	C098088	45/F	10400	64	29	5	2	10	16	89	119	30	0.9	33	32	174	151	NIL	NIL	1-2	1-2
5	C99770	20/F	7600	61	33	4	2	14	20	93	123	29	0.7	32	28	199	163	NIL	NIL	1-2	1-2
6	C97890	65/F	6800	54	43	2	1	8	12	103	133	27	0.8	30	29	256	157	NIL	NIL	2-3	2-3
7	C97884	23/M	7200	58	35	5	2	14	22	95	125	30	0.8	33	32	212	169	NIL	NIL	1-2	1-2
8	C89948	45/F	8400	67	28	4	1	8	14	88	118	29	0.6	32	31	167	146	NIL	NIL	2-3	1-2
9	C90117	45/F	9100	72	26	2	0	6	12	86	116	27	0.7	30	27	156	140	NIL	NIL	2-3	2-3
10	D00452	62/M	7900	63	32	4	1	12	16	92	122	29	0.9	32	31	191	154	NIL	NIL	1-2	1-2
11	C98655	34/F	10500	67	32	1	0	10	18	92	122	26	1	29	25	192	158	NIL	NIL	1-2	2-3
12	C91131	43/F	8800	70	26	3	1	6	10	86	116	28	0.6	31	30	154	136	NIL	NIL	1-2	1-2
13	C9941	45/F	9400	67	29	3	1	10	18	89	119	28	0.8	31	26	174	155	NIL	NIL	1-2	2-3
14	C00347	46/F	7600	61	32	5	2	14	20	92	122	30	1	33	32	193	162	NIL	NIL	2-3	1-2
15	C95755	30/F	9900	69	28	2	1	8	12	88	118	27	0.9	30	27	168	142	NIL	NIL	1-2	2-3
16	C67253	38/F	10200	71	25	3	1	6	10	85	115	28	1	31	30	150	135	NIL	NIL	1-2	2-3
17	C002479	48/F	10100	62	37	1	0	8	12	97	127	26	0.7	29	28	222	151	NIL	NIL	1-2	2-3
18	C90388	35/F	9300	73	20	5	2	14	20	80	110	30	0.9	33	26	120	150	NIL	NIL	2-3	2-3
19	C001189	33/F	8200	66	29	4	1	12	20	89	119	29	0.6	32	29	176	159	NIL	NIL	1-2	1-2
20	C97754	49/F	9700	71	25	3	1	8	16	85	115	28	0.8	31	28	150	147	NIL	NIL	1-2	2-3

LAB INVESTINGATIONS – AFTER TREATMENT ANAEMIA PATIENTS																					
SL No	OP No	Age/Sex	TC	Dc (in %)				ESR (mm/hr)		BS (mg/dl)		Urea	Creat	OT	PT	Al.P	T.Chol	Urine			
				P	L	E	M	1/2	1	F	PP/R							Alb	Sug	Dep	
																				Pus cells	Epi cells
1	C83474	50/F	7400	62	37	1	0	12	26	100	130	26	0.7	29	26	222	192	NIL	NIL	1-2	2-3
2	C85480	60/F	8400	69	28	2	1	10	16	93	123	28	0.5	31	26	178	151	NIL	NIL	1-2	1-2
3	C85924	42/F	9500	69	30	1	0	10	16	89	119	26	0.8	29	28	158	157	NIL	NIL	2-3	2-3
4	C098088	45/F	10200	68	25	5	2	8	14	94	124	32	1	35	30	166	146	NIL	NIL	1-2	1-2
5	C99770	20/F	7700	58	36	4	2	8	12	97	127	31	0.7	34	26	191	159	NIL	NIL	1-2	1-2
6	C97890	65/F	6700	53	44	2	1	2	4	105	135	28	0.8	31	28	254	155	NIL	NIL	2-3	2-3
7	C97884	23/M	7400	62	31	5	2	10	18	100	130	32	0.7	35	30	202	164	NIL	NIL	1-2	1-2
8	C89948	45/F	8700	69	26	4	1	8	12	92	122	30	0.6	33	30	159	142	NIL	NIL	2-3	1-2
9	C90117	45/F	8900	70	28	2	0	4	6	88	118	27	0.7	30	27	152	138	NIL	NIL	2-3	2-3
10	D00452	62/M	8000	61	34	4	1	6	10	96	126	30	0.9	33	30	185	150	NIL	NIL	1-2	1-2
11	C98655	34/F	10400	71	28	1	0	4	10	93	123	26	1	29	25	188	157	NIL	NIL	1-2	2-3
12	C91131	43/F	9000	67	29	3	1	2	4	89	119	29	0.6	32	29	152	133	NIL	NIL	1-2	1-2
13	C9941	45/F	9700	66	30	3	1	8	14	92	122	29	0.8	32	25	166	152	NIL	NIL	1-2	2-3
14	C00347	46/F	7400	65	28	5	2	6	14	97	127	32	1	35	30	187	157	NIL	NIL	2-3	1-2
15	C95755	30/F	10000	71	26	2	1	2	4	90	120	28	0.9	31	26	166	140	NIL	NIL	1-2	2-3
16	C67253	38/F	10100	74	22	3	1	6	10	88	118	29	1	32	29	144	132	NIL	NIL	1-2	2-3
17	C002479	48/F	10300	60	39	1	0	4	6	98	128	26	0.8	29	28	218	150	NIL	NIL	1-2	2-3
18	C90388	35/F	9600	71	22	5	2	6	12	85	115	32	0.7	35	24	114	145	NIL	NIL	2-3	2-3
19	C001189	33/F	8000	63	32	4	1	10	16	93	123	30	0.6	33	28	166	155	NIL	NIL	1-2	1-2
20	C97754	49/F	9800	70	26	3	1	2	12	88	118	29	0.7	32	27	148	144	NIL	NIL	1-2	2-3

LAB INVESTIGATIONS - ANAEMIA																
SL NO	OP No	Name	Hb (gm%)		TRbc (millions/ μ l)		HCT/PCV (%)		MCV (fl)		MCH (pg)		MCHC (gm/dl)		Smear Study	
			BT	AT	BT	AT	BT	AT	BT	AT	BT	AT	BT	AT	BT	AT
1	C83474	Mrs.V.Padma	7.5	8.3	3.8	3.8	28.9	29	70.4	70.5	21.1	22	29.7	29.8	+	+
2	C85480	Mrs.R.Arputham	9.6	10.2	4.1	4.1	29	30	71.2	71	22.1	23	28	29	+	-
3	C85924	Mrs.Dhatchayini	8.5	9.2	4.2	4.2	30	30.1	69.1	70	20.3	21	29.1	29	+	+
4	C098088	Mrs.C.Athilakshmi	9.3	10.2	4	4.1	28.4	29	72	72.1	21	20.6	28.3	28.4	+	-
5	C99770	Miss.S.Sudha	7.2	8	4	4.1	24.6	25	71.9	72	24.1	25	27.9	28	+	+
6	C97890	Mrs.P.Saratha	7.2	8	3.9	4	26	26.1	65.4	66	23.2	24	28.1	28	+	+
7	C97884	Mr.M.Ravi	7	7.8	3.8	3.9	25	25.1	68.3	69	22.8	23	28.1	28.9	+	+
8	C89948	Mrs.R.Panjali	9.9	10.5	3.8	3.8	32	32.1	66.9	67	23.1	24	29.3	30.1	+	-
9	C90117	Mrs.K.G.Govindammal	9.7	10.4	4.1	4.1	33	33.2	69.9	69.8	22	23	30.4	31	+	-
10	D00452	Mr.P.Loganathan	9.2	10.4	3.9	4	31.1	32	69.8	70	20	21	29.8	30	+	-
11	C98655	Mrs.K.S.Rani	9.8	10.6	4.4	4.4	32.5	32	67.3	68	21.3	22	28.1	29	+	-
12	C91131	Mrs.S.Mythili	9	10.2	3.9	3.9	31	31.1	65.9	66	21.1	21.4	28.3	29.9	+	-
13	C9941	Mrs.Lally Baskaran	9	10.1	4	4	30	31.2	66	67	23.9	24.1	28.3	28.4	+	-
14	C00347	Mrs.I.Banu	8.8	9.8	4.1	4.1	29.6	30.4	65.2	66	24	24.4	29.3	29.9	+	+
15	C95755	Mrs.S.Maruthi	7.6	8.8	4.2	4.2	29	30	69	69.3	19.8	20	28.5	29	+	+
16	C67253	Mrs.R.Malliga	8.6	9.6	4.3	4.3	31.4	32	68	68.5	19.9	21	27	27.5	+	+
17	C002479	Mrs.K.Gomathi	9.2	10.4	4.1	4.1	32.3	33	69.4	70	20	21	27	27	+	-
18	C90388	Mrs.A.Shanthi	7.6	9.2	4	4	25.9	26	70.8	71	19.7	20.3	27.4	27.8	+	+
19	C001189	Mrs.L.Shoba	9.1	10	3.9	3.9	30	30.2	69.1	69	18	19	28.1	28	+	+
20	C97754	Mrs.M.Muthulakshmi	7.8	8.6	3.7	3.8	28	28.2	68.5	69	19	19.3	29.5	29.6	+	+

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